ALBT2

cobas®

Tina-quant Albumin CSF

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08056722190	Tina-quant Albumin Gen.2 (500 tests)	System-ID 2006 001	**cobas c** 303, **cobas c** 503
Materials required (but not provided):			
03121305122	C.f.a.s. PUC (5 x 1 mL)	Code 20489	
03121291122	Precipath PUC (4 x 3 mL)	Code 20241	
10557897122	Precinorm Protein (3 x 1 mL)	Code 20302	
11333127122	Precipath Protein (3 x 1 mL)	Code 20303	
05117003190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	
08059322190	Antigen Excess Reagent (START) (1000 tests)	System-ID 2008 001	

English

System information

ALBT2C: ACN 20062 (Albumin in CSF)

ALBT2R: ACN 20067 (Albumin in CSF, application in serum/plasma)

Intended use

In vitro test for the quantitative determination of albumin in human cerebrospinal fluid and corresponding human serum/plasma on Roche/Hitachi **cobas c** systems.

Summary[1,2,3,4,5,6,7]

Cerebrospinal fluid (CSF) analysis is a basic tool for diagnosis of neurological diseases.
The diffusion of proteins through the blood-brain barrier normally occurs at a steady rate. The rate is influenced by the permeability of the blood-brain barrier and CSF flow rate. Changes in protein concentration in the CSF can be an indication for various neurological diseases.
Disease-related immunoglobulin patterns (IgG, IgA, IgM with reference to albumin) allow for the differential diagnosis of neurological disorders with the aid of Reiber quotient schemes.
Elevated levels of IgG, IgA, IgM in CSF are often associated with opportunistic infections of the central nervous system (CNS) and neurotuberculosis. Increased CSF IgG, IgA, IgM concentrations may occur because of either increased permeability of the blood-brain barrier or local/intrathecal production of IgG, IgA, IgM or both. Malfunction of the blood-brain barrier can be reliably quantified by means of the albumin CSF/serum ratio.
Albumin is an ideal reference protein for blood-brain barrier function, since it is solely synthesized outside the brain and thereby provides an excellent measure for proteins passing the blood-brain barrier. An elevated albumin CSF/serum ratio is an indication of disorders of the blood-brain barrier.
Measuring IgG, IgA, IgM and albumin in CSF/serum pairs, a differentiation between IgG, IgA, IgM originating from blood and IgG, IgA, IgM originating from intrathecal production is possible. The results of the CSF/serum ratio for IgG, IgA, IgM and Albumin, in conjunction with Reiber quotient scheme provide an aid in the diagnosis of functional blood-brain barriers disorders and/or intrathecal IgG, IgA, IgM synthesis.
Blood brain barrier disorders can be reliably quantified with the aid of the albumin CSF/serum ratio. Elevated albumin ratios are indicative of a blood-brain barrier disorder.
By simultaneously determining IgG, IgA, IgM in CSF and serum while taking into account the individual albumin ratios, it is possible to differentiate between IgG, IgA, IgM originating from the blood and CNS-synthesized immunoglobulin.
Albumin is a non-glycosylated protein with a molecular weight of 66000 Da. It is synthesized in liver parenchymal cells at a rate of 14 g/day.
Quantitatively, albumin is normally the most important protein component (> 50 %) in plasma, CSF and urine.
Albumin has two main functions in plasma: maintaining the oncotic pressure (80 % due to albumin in plasma) and transport. It is the most important transport protein for substances having low water solubility (such as free fatty acids, bilirubin, metal ions, hormones and pharmaceuticals).
Depressed albumin levels are caused by hyperhydration, hepatocellular synthesis insufficiency, secretion disorders in the intravascular space, abnormal distribution between the intravascular and extravascular space, catabolism and loss of albumin, acute phase reactions and congenital analbuminemia.

Test principle

Immunoturbidimetric assay
Anti-albumin antibodies react with the antigen in the sample to form antigen/antibody complexes which, following agglutination, are measured turbidimetrically.

Reagents - working solutions

R1 TRIS buffer: 50 mmol/L, pH 8.0; PEG: ≥ 4.2 %; EDTA: 2.0 mmol/L; preservative

R2 Polyclonal anti-human albumin antibodies (sheep): dependent on titer; TRIS buffer: 100 mmol/L, pH 7.2; preservative

R1 is in position B and R2 is in position C.

Antigen excess reagent (Cat. No. 08059322190, for ACN 20062):

R3 Diluted serum (human); NaCl: 150 mmol/L; phosphate buffer: 50 mmol/L, pH 7.0; preservative

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.

All human material should be considered potentially infectious. All products derived from human blood are prepared exclusively from the blood of donors tested individually and shown to be free from HBsAg and antibodies to HCV and HIV. The testing methods use assays that have been approved by the FDA or that are in compliance with the legal rules applicable to placing in vitro diagnostic medical devices for human use on the market in the European Union.
However, as no testing method can rule out the potential risk of infection with absolute certainty, the material should be handled with the same level of care as a patient specimen. In the event of exposure, the directives of the responsible health authorities should be followed.[8,9]

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.

ALBT2

Tina-quant Albumin CSF

cobas®

On-board in use and refrigerated on the analyzer: 26 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.
Cerebrospinal fliud (CSF)
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.

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

Stability in CSF:[10]	up to 3 days at 2-8 °C
	6 months at (–15)-(–25) °C
	indefinitely at (–60)-(–80) °C
Stability in serum/plasma:[11]	10 weeks at 15-25 °C
	5 months at 2-8 °C
	4 months at (–15)-(–25) °C

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 CSF

Test definition

Reporting time	10 min	
Wavelength (sub/main)	700/340 nm	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	–
R2	16 µL	–
R3	5 µL	16 µL

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	4.8 µL	10 µL	110 µL
Decreased	2.4 µL	2.5 µL	90 µL
Increased	4.8 µL	10 µL	110 µL

Application for serum and plasma

Test definition

Reporting time	10 min	
Wavelength (sub/main)	700/340 nm	
Reagent pipetting		Diluent (H_2O)
R1	80 µL	–
R2	16 µL	–

Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.6 µL	1.5 µL	125 µL
Decreased	1.6 µL	1.0 µL	106 µL
Increased	1.6 µL	1.5 µL	125 µL

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

Calibration

Application for CSF (ACN 20062)

Calibrators	S1: H_2O
	S2-6: C.f.a.s. PUC
Calibration mode	Non-linear
Calibration frequency	Full calibration - after reagent lot change - after 12 weeks on-board - as required following quality control procedures

Application for serum/plasma (ACN 20067)
Transfer of calibration from CSF application (ACN 20062)

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

Traceability: This method has been standardized against the certified reference material in human serum of the IRMM (Institute for Reference Materials and Measurements) ERM-DA470k/IFCC.

Quality control

For quality control, use control materials as listed in the "Order information" section. In addition, other suitable control material can be used.

CSF	undiluted Precipath PUC
Serum/plasma	Precinorm Protein, Precipath Protein, PreciControl ClinChem Multi 1, PreciControl ClinChem Multi 2

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 (mg/dL, g/L).

Conversion factors:	mg/L × 0.1 = mg/dL
	mg/L × 0.001 = g/L

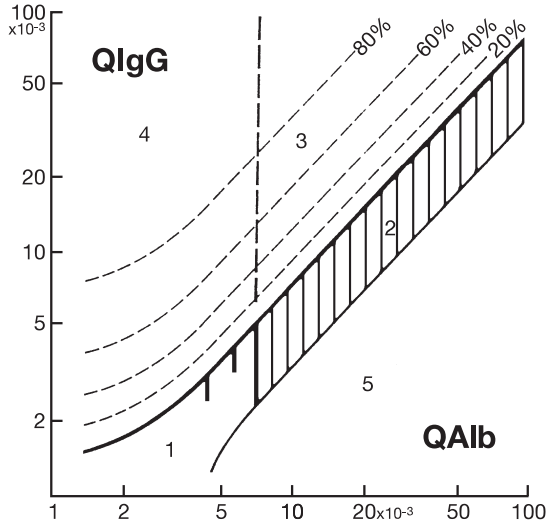
Reiber Quotient Graph

With the aid of commercially available software, Reiber Quotient Diagrams can be automatically generated.

The calculation employs a ratio diagram including hyperbolic functions as differential lines according to Reiber and Felgenhauer. Results from the determination of IgG and albumin in CSF and serum (IgG and albumin ratios)[12] are plotted.

ALBT2

Tina-quant Albumin CSF



1. Reference range. 2. Blood brain barrier functional disorder without local IgG synthesis. 3. Blood brain barrier functional disorder with concomitant IgG-synthesis in the CNS. 4. IgG synthesis in the CNS without blood brain barrier functional disorder. 5. As confirmed empirically, there are no values in this region (i.e. values here are due to errors introduced by blood sampling or analytical errors). Generally speaking, cases not associated with local IgG synthesis in the CNS lie below the bold line (hyperbolic function). The percentage values indicate what percentage of the total IgG in CSF (minimum) originates in the CNS relative to the statistically-defined 0 % differential lines.

Limitations - interference

CSF

Criterion: Recovery within ± 10 % of initial value at an albumin concentration of 240 mg/L.

Icterus: No significant interference up to an I index of 60 for conjugated bilirubin (approximate conjugated bilirubin concentration: 1026 µmol/L or 60 mg/dL).

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

High dose hook-effect: Using the prozone check automatically performed by the analyzer, no false result without a flag was observed up to an albumin concentration of 30000 mg/L.

Serum/plasma

Criterion: Recovery within ± 10 % of initial value at an albumin concentration of 35 g/L.

Icterus:[13] 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:[13] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[13] No significant interference up to an L index of 1500. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

Rheumatoid factors: No significant interference up to a concentration of 1200 IU/mL.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[14,15]

In very rare cases, gammopathy, in particular type IgM (Waldenström's macroglobulinemia), may cause unreliable results.[16]

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

CSF

36-4800 mg/L

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:6.2 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 6.2.

Serum, plasma

3-101 g/L

Determine samples having higher concentrations via the rerun function. Dilution of samples via the rerun function is a 1:1.27 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 1.27.

Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation
CSF

Limit of Blank	= 20 mg/L
Limit of Detection	= 36 mg/L
Limit of Quantitation	= 50 mg/L

Serum, plasma

Limit of Blank	= 1 g/L
Limit of Detection	= 3 g/L
Limit of Quantitation	= 3 g/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 20 %. It has been determined using low concentration albumin samples.

Expected values

Serum/plasma

Consensus values:[17]

Adults	35-52 g/L

Reference intervals according to Tietz:[18]

Newborns (0-4 days):	28-44 g/L
Children (4 days-14 years):	38-54 g/L

Albumin CSF/serum ration (Q_{ALB} × 10^3)

Adults[3]	up to 15 years	5.0
	up to 40 years	6.5
	up to 60 years	8.0

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

ALBT2

Tina-quant Albumin CSF

cobas®

Specific performance data

Representative performance data on the analyzers are given below. These data represent the performance of the analytical procedure itself.

Results obtained in individual laboratories may differ due to heterogenous sample materials, aging of analyzer components and mixture of reagents running on the analyzer.

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.

CSF

Repeatability	*Mean mg/L*	*SD mg/L*	*CV %*
Precipath PUC	117	2.32	2.0
CSF 1	108	2.78	2.6
CSF 2	226	2.93	1.3
CSF 3	338	2.81	0.8
CSF 4	2256	18.1	0.8
CSF 5	3911	42.8	1.1

Intermediate precision	*Mean mg/L*	*SD mg/L*	*CV %*
Precipath PUC	121	2.78	2.3
CSF 1	108	3.28	3.0
CSF 2	232	5.95	2.6
CSF 3	340	6.07	1.8
CSF 4	2226	33.9	1.5
CSF 5	3911	96.7	2.5

Serum/plasma

Repeatability	*Mean g/L*	*SD g/L*	*CV %*
PCCC1[a)]	30.9	0.290	0.9
PCCC2[b)]	44.2	0.371	0.8
Serum 1	5.10	0.0552	1.1
Serum 2	24.5	0.309	1.3
Serum 3	39.1	0.327	0.8
Serum 4	49.3	0.397	0.8
Serum 5	80.4	1.02	1.3

Intermediate precision	*Mean g/L*	*SD g/L*	*CV %*
PCCC1[a)]	30.4	0.376	1.2
PCCC2[b)]	44.2	0.573	1.3
Serum 1	5.10	0.0595	1.2
Serum 2	24.5	0.385	1.6
Serum 3	39.1	0.458	1.2
Serum 4	49.3	0.467	0.9
Serum 5	80.4	1.07	1.3

a) PreciControl ClinChem Multi 1

b) PreciControl ClinChem Multi 2

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

Method comparison

Albumin values for human CSF, serum and plasma 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).

CSF

Sample size (n) = 75

Passing/Bablok[19]	Linear regression
y = 0.970x + 11.3 mg/L	y = 0.937x + 25.0 mg/dL
τ = 0.988	r = 1.000

The sample concentrations were between 43.4 and 4634 mg/L.

Serum/plasma

Sample size (n) = 1306

Passing/Bablok[19]	Linear regression
y = 0.995x + 1.06 g/L	y = 0.995x + 1.18 g/L
τ = 0.912	r = 0.995

The sample concentrations were between 4.20 and 101 g/L.

Albumin values for human CSF, serum and plasma 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).

CSF

Sample size (n) = 75

Passing/Bablok[19]	Linear regression
y = 1.019x + 1.61 mg/L	y = 0.991x + 15.2 mg/L
τ = 0.986	r = 1.000

The sample concentrations were between 42.0 and 4545 mg/L.

Serum/plasma

Sample size (n) = 74

Passing/Bablok[19]	Linear regression
y = 0.984x + 1.30 g/L	y = 0.963x + 1.97 g/L
τ = 0.950	r = 0.999

The sample concentrations were between 3.96 and 100 g/L.

References

1 Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie, 3rd ed. Stuttgart/New York: Schattauer Verlag 1995:749-750.

2 Rothschild MA, Oratz M, Schreiber SS. Serum albumin. Hepatology 1988;8(2):385-401.

3 Reiber H. External Quality Assessment in Clinical Neurochemistry: Survey of Analysis for Cerebrospinal Fluid (CSF) Proteins based on CSF/Serum Quotients. Clin Chem 1995;41:256-263.

4 Reiber H, Felgenhauer K. Protein transfer of the blood cerebrospinal fluid barrier and the quantitation of the humoral immune response within the central nervous system. Clin Chim Acta 1987;163(3):319-328.

5 Reiber H. Die diagnostische Bedeutung neuroimmunologischer Reaktionsmuster im Liquor cerebrospinalis. Lab med 1995;19:444-462.

6 Zimmermann K, Marr U, Linke E. Liquordiagnostik, MTA 1996;11:258-260.

7 Reiber H. Flow rate of cerebrospinal fluid (CSF) – a concept common to normal blood-CSF barrier function and to dysfunction in neurological diseases. J Neurol Sci 1994;122:189-203.

8 Occupational Safety and Health Standards: Bloodborne pathogens. (29 CFR Part 1910.1030). Fed. Register.

9 Directive 2000/54/EC of the European Parliament and Council of 18 September 2000 on the protection of workers from risks related to exposure to biological agents at work.

10 Tietz NW, ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: WB Saunders Company 1995;24.

11 Use of Anticoagulants in Diagnostic Laboratory Investigations. WHO Publication WHO/DIL/LAB/99.1 Rev. 2: Jan 2002.

12 Reiber H. The hyperbolic function: a mathematical solution of the protein flux/CSF flow model for blood-CSF barrier function. J Neurol Sci 1994;126:243-245.

13 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

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

15 Sonntag O, Scholer A. Drug interference in clinical chemistry: recommendation of drugs and their concentrations to be used in drug interference studies. Ann Clin Biochem 2001;38:376-385.

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

17 Dati F, Schumann G, Thomas L, et al. Consensus of a group of professional societies and diagnostic companies on guidelines for interim reference ranges for 14 proteins in serum based on the standardization against the IFCC/BCR/CAP reference material (CRM 470). Eur J Clin Chem Clin Biochem 1996;34:517-520.

18 Wu AHB, ed. Tietz Clinical Guide to Laboratory Tests, 4th ed. WB Saunders Company, 2006:66.

19 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.

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 after reconstitution or mixing
GTIN	Global Trade Item Number

CE 0123

Roche Diagnostics GmbH, Sandhofer Strasse 116, D-68305 Mannheim
www.roche.com
+800 5505 6606