AAGP2

cobas®

Tina-quant α1-Acid Glycoprotein Gen.2

Order information

REF	CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
03333795190	Tina-quant α1-Acid Glycoprotein Gen.2 (100 tests)	System-ID 07 6758 1	COBAS INTEGRA 400 plus

Materials required (but not provided):

11355279216	Calibrator f.a.s. Proteins (5 × 1 mL)	System-ID 07 6557 0	
11355279160	Calibrator f.a.s. Proteins (5 × 1 mL, for USA)	System-ID 07 6557 0	
10557897122	Precinorm Protein (3 × 1 mL)	System-ID 07 9105 9	
10557897160	Precinorm Protein (3 × 1 mL, for USA)	System-ID 07 9105 9	
11333127122	Precipath Protein (3 × 1 mL)	System-ID 07 9106 7	
11333127160	Precipath Protein (3 × 1 mL, for USA)	System-ID 07 9106 7	
05117003190	PreciControl ClinChem Multi 1 (20 × 5 mL)	System-ID 07 7469 3	
05947626190	PreciControl ClinChem Multi 1 (4 × 5 mL)	System-ID 07 7469 3	
05947626160	PreciControl ClinChem Multi 1 (4 × 5 mL, for USA)	System-ID 07 7469 3	
05117216190	PreciControl ClinChem Multi 2 (20 × 5 mL)	System-ID 07 7470 7	
05947774190	PreciControl ClinChem Multi 2 (4 × 5 mL)	System-ID 07 7470 7	
05947774160	PreciControl ClinChem Multi 2 (4 × 5 mL, for USA)	System-ID 07 7470 7	
20756350322	NaCl Diluent 9 % (6 × 22 mL)	System-ID 07 5635 0	

English

System information

Test AAGP2, test ID 0-258

Intended use

In vitro test for the quantitative immunological determination of human α_1-acid glycoprotein in human serum and plasma on COBAS INTEGRA systems.

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

α_1-Acid glycoprotein is synthesized in hepatocytes and consists of a polypeptide chain having 5 carbohydrate chains N-glycosidically bonded to it (molar mass 41000 daltons). Structurally, it belongs to the lipocalin superfamily of secretory proteins (such as α_1-microglobulin and retinol-binding protein). α_1-Acid glycoprotein promotes fibroblast growth and interacts with collagen.

It is a sensitive acute phase reactant whose concentration can increase by a factor of 3 within 24-48 hours when inflammation occurs. α_1-Acid glycoprotein can also be used to differentiate between acute phase reactions (elevated serum level) and estrogen effects (normal or decreased serum level) whereas the serum level of other positive reactants such as ceruloplasmin and haptoglobin increases during such reactions. Along with haptoglobin it is perhaps the best protein for identifying slight in vivo hemolysis. An increased α_1-acid glycoprotein level and normal haptoglobin values indicate an acute phase reaction with concomitant slight in vivo hemolysis. Moderate and isolated increases occur when glomerular filtration is inhibited in the early stages of uremia. The determination is used in the assessment of the activity of acute and recurring inflammations as well as of tumors with cell necrosis.

Various assay methods for α_1-acid glycoprotein determination are available such as kinetic nephelometry, radial immunodiffusion (RID) and turbidimetry. The Roche α_1-acid glycoprotein assay is based on the principle of immunological agglutination.

Test principle[5]

Immunoturbidimetric assay

- Sample and addition of R1
- Addition of SR and start of reaction

Anti-α_1-acid glycoprotein antibodies react with antigen in the sample to form an antigen/antibody complex. Following agglutination, this is measured turbidimetrically.

Reagents - working solutions

R1 TRIS buffer: 50 mmol/L, pH 8.0; PEG: 7 %; NaCl: 300 mmol/L; preservative; stabilizer

SR Polyclonal anti-human α_1-acid glycoprotein antibody (goat): dependent on titer; TRIS buffer: 13 mmol/L, pH 7.5; NaCl: 198 mmol/L; preservative

R1 is in position B and SR 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.

For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

Reagent handling

Ready for use

Storage and stability

Shelf life at 2-8 °C	See expiration date on **cobas c** pack label
COBAS INTEGRA 400 plus system	
On-board in use at 10-15 °C	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-, Na-, NH_4^+-heparin; K_2-, K_3-EDTA

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.

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

Centrifuge samples containing precipitates before performing the assay.

Stability:[6]	< 72 hours at 4 °C
	6 months at –20 °C

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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)

NaCl Diluent 9 %, Cat. No. 20756350322, system-ID 07 5635 0 for automatic sample dilution and standard serial dilutions. NaCl Diluent 9 % is placed in its predefined rack position and is stable for 4 weeks on-board COBAS INTEGRA 400 plus analyzers.

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.

Application for serum and plasma

COBAS INTEGRA 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	33/69
Typical prozone effect	> 11.0 g/L (> 1100 mg/dL or > 275 µmol/L)
Antigen excess check	No
Predilution factor	21
Unit	g/L

Pipetting parameters

		Diluent (H_2O)
R1	120 µL	
Sample	12 µL	8 µL
SR	40 µL	8 µL
Total volume	188 µL	

Calibration

Calibrator	C.f.a.s. Proteins
Calibration dilution ratio	1:7, 1:15, 1:30, 1:75, and 0 g/L performed automatically by the instrument.
Calibration mode	Logit/log 4
Calibration replicate	Duplicate recommended
Calibration interval	Each lot and as required following quality control procedures

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

Enter the assigned lot-specific α_1-acid glycoprotein value of the undiluted calibrator indicated in the package insert for C.f.a.s. Proteins.

Traceability: This method is standardized against an internal method traceable to CRM 470.

Quality control

Reference range	Precinorm Protein or PreciControl ClinChem Multi 1
Pathological range	Precipath Protein or PreciControl ClinChem Multi 2
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. 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

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 plus analyzer).

Conversion factors:	g/L × 25 = µmol/L
	g/L × 100 = mg/dL
	mg/dL × 0.25 = µmol/L

Limitations - interference

Criterion: Recovery within ± 10 % of initial value.

Icterus:[7] No significant interference up to an I index of 60 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 60 mg/dL or 1026 µmol/L).

Hemolysis:[7] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 1000 mg/dL or 621 µmol/L).

Lipemia:[7] No significant interference up to an L index of 700. 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.

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 COBAS INTEGRA analyzers. Refer to the CLEAN Method Sheet for further instructions and for the latest version of the Extra wash cycle list.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

0.25-4.0 g/L (6.25-100 µmol/L or 25-400 mg/dL) (typical measuring range)

The upper limit of the measuring range depends on the actual calibrator value.

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 using the rerun function are automatically multiplied by a factor of 2.

Lower limits of measurement

Lower detection limit of the test:
0.10 g/L (2.5 µmol/L or 10 mg/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 a zero sample (zero sample + 3 SD, repeatability, n = 30).

Expected values*

0.5-1.2 g/L (50-120 mg/dL or 12.5-30 µmol/L)

* Reference range according to CRM 470 protein standardization[11]

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 COBAS INTEGRA 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 (1 aliquot per run, 1 run per day, 21 days). The following results were obtained:

Repeatability	*Mean g/L (µmol/L)*	*SD g/L (µmol/L)*	*CV %*
Precinorm Protein	0.83 (20.8)	0.01 (0.3)	1.8
Precipath Protein	1.39 (34.8)	0.02 (0.5)	1.5
Human serum	0.87 (21.8)	0.02 (0.5)	2.0

Intermediate precision	*Mean g/L (µmol/L)*	*SD g/L (µmol/L)*	*CV %*
Precinorm Protein	0.80 (20.0)	0.02 (0.5)	2.3
Precipath Protein	1.34 (33.5)	0.02 (0.5)	1.7
Human serum	0.85 (21.3)	0.02 (0.5)	2.9

Method comparison

α_1-Acid glycoprotein values for human serum and plasma samples obtained on a COBAS INTEGRA 700 analyzer with the COBAS INTEGRA AAGP Gen.2 reagent (y) were compared with those determined using the same reagent on a Roche/Hitachi 917 analyzer (x) and with the previous reagent (AAGP) on a COBAS INTEGRA 700 analyzer (x).

Roche/Hitachi 917 analyzer	Sample size (n) = 55
Passing/Bablok[12]	Linear regression
y = 1.043x - 0.043 g/L	y = 1.027x - 0.025 g/L
τ = 0.945	r = 0.996
SD (md 95) = 0.074	Sy.x = 0.041

The sample concentrations were between 0.4 and 2.85 g/L (40 and 285 mg/dL or 10 and 71.3 µmol/L).

COBAS INTEGRA 700 analyzer	Sample size (n) = 55
Passing/Bablok[12]	Linear regression
y = 1.005x - 0.005 g/L	y = 1.001x + 0.001 g/L
τ = 0.964	r = 0.998
SD (md 95) = 0.056	Sy.x = 0.027

The sample concentrations were between 0.4 and 2.75 g/L (40 and 275 mg/dL or 10 and 68.8 µmol/L).

References

1 Schmid K. α1-Acid glycoprotein. In: The Plasma Proteins, 2nd ed. Putnam FW, ed. New York: Academic Press 1975;183-228.

2 Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie, 3rd ed. Stuttgart/New York: Schattauer Verlag 1995:234-236.

3 Tietz NW, ed. Clinical Guide to Laboratory Tests, 3rd ed. Philadelphia, PA: WB Saunders Company 1995;66-67.

4 Ganrot K. Plasma protein pattern in acute infectious disease. Scand J Clin Lab Invest 1974;34:75-81.

5 Lievens M, Bienvenu J, Buitrago JMG, et al. Evaluation of four new Tina-quant assays for determination of α1-acid glycoprotein, α1-antitrypsin, haptoglobin and prealbumin. Clin Lab 1996;42:515-520.

6 Wu AHB, ed. Tietz Clinical Guide to Laboratory Tests, 4th ed. St. Louis (MO): Saunders Elsevier 2006:42.

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

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

9 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.

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 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.

12 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 for reconstitution
GTIN	Global Trade Item Number

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