ASLOT

cobas®

Tina-quant Antistreptolysin O

Order information

REF		CONTENT		Analyzer(s) on which **cobas c** pack(s) can be used
08105472190	08105472500	Tina-quant Antistreptolysin O (200 tests)	System-ID 2021 001	**cobas c** 303, **cobas c** 503, **cobas c** 703

Materials required (but not provided):

03555941190	C.f.a.s. PAC (3 × 1 mL)	Code 20589	
10557897122	Precinorm Protein (3 × 1 mL)	Code 20302	
11333127122	Precipath Protein (3 × 1 mL)	Code 20303	
05117003190	PreciControl ClinChem Multi 1 (20 × 5 mL)	Code 20391	
05947626190	PreciControl ClinChem Multi 1 (4 × 5 mL)	Code 20391	
05117216190	PreciControl ClinChem Multi 2 (20 × 5 mL)	Code 20392	
05947774190	PreciControl ClinChem Multi 2 (4 × 5 mL)	Code 20392	
08063494190	Diluent NaCl 9 % (123 mL)	System-ID 2906 001	

English

System information

ASLOT: ACN 20210

Intended use

In vitro test for the quantitative immunological determination of antistreptolysin O in human serum and plasma on **cobas c** systems.

Summary

Antistreptolysin O (ASO) measurements, performed with this assay in human serum and plasma, are used as an aid in the diagnosis of antecedent group A streptococcus infection, which can be associated with post-infectious complications.

Group A streptococcus (GAS; Streptococcus pyogenes) is a Gram-positive, β-haemolytic bacterium which most commonly infects the throat or skin.[1,2] The ability of GAS to overcome innate and acquired immune mechanisms present in saliva allows the bacterium to remain viable for a long period.[1] Severe GAS infection results from the ability of the bacterium to migrate to normally sterile sites, such as the bloodstream and deep tissues.[1] Here, the interaction between host and pathogen factors leads to tissue destruction, bacterial dissemination and hyperinflammation.[1] Immunological defense reactions can be induced by several metabolites of the β-hemolyzing streptococci, which act as exogenous toxins for the human organism.[1] The most clinically important antibody reactions are found against streptolysin O, streptococcal deoxyribonuclease B, hyaluronidase and streptokinase. Determination of the antistreptolysin O antibody level is widely adopted to obtain useful information on preceding streptococcal infection.[2,3]

GAS is the cause of a wide range of acute, common pyogenic infections, including skin diseases or tonsillopharyngitis that may be followed by non-suppurative complications including acute rheumatic fever (ARF), rheumatic heart disease (RHD), poststreptococcal glomerulonephritis (PSGN), poststreptococcal reactive arthritis (PSRA) and pediatric autoimmune neuropsychiatric disorder associated with streptococcal infection (PANDAS).[2,3,4,5,6,7]

Early diagnosis, efficient treatment and monitoring of the patient can reduce risks and aid in management of post-infection complications.[8] Antistreptococcal antibody titers reflect past immunologic events. Antistreptolysin O titers begin to rise approximately 1 week and peak 3 to 6 weeks after the infection.[5] Ideally, to optimize diagnosis of preceding GAS infection, 2 sequential ASO measurements should be performed.[2,9] A fourfold or greater rise between 2 successive serological samples (10-14 days apart) in ASO titer is indicative of recent GAS infection.[9]

Test principle[10,11,12,13]

Immunoturbidimetric assay

Human antistreptolysin O antibodies agglutinate with latex particles coated with streptolysin O antigens. The precipitate is determined turbidimetrically.

Reagents - working solutions

R1 TRIS buffer: 170 mmol/L, pH 8.2

R3 Borate buffer: 10 mmol/L, pH 8.2; latex particles coated with streptolysin O: 2 mL/L

R1 is in position B and 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.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Danger

H317	May cause an allergic skin reaction.
H360FD	May damage fertility. May damage the unborn child.
H412	Harmful to aquatic life with long lasting effects.

Prevention:

P201	Obtain special instructions before use.
P261	Avoid breathing mist or vapours.
P273	Avoid release to the environment.
P280	Wear protective gloves/ protective clothing/ eye protection/ face protection/ hearing protection.

Response:

P308 + P313	IF exposed or concerned: Get medical advice/attention.
P333 + P313	If skin irritation or rash occurs: Get medical advice/attention.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590

Reagent handling

Ready for use

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Carefully invert reagent container several times prior to use to ensure that the reagent components are mixed.

Storage and stability

Shelf life at 2-8 °C:	See expiration date on **cobas c** pack label.
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.
Serum
Plasma: Li-heparin and K_2-EDTA plasma

The use of plasma can lead to a decrease in antistreptolysin O activity of approximately 7 %. For samples with an activity below 100 IU/mL the recovery in plasma can be either decreased or increased in comparison to serum.

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:[14]	2 days at 20-25 °C
	8 days at 4-8 °C
	6 months at -20 °C (±5 °C)

Freeze only once.

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

Test definition

Reporting time	10 min		
Wavelength (sub/main)	–/700 nm		
Reagent pipetting		Diluent (H_2O)	
R1	63 µL	–	
R3	63 µL	–	
Sample volumes	*Sample*	*Sample dilution*	
		Sample	*Diluent (NaCl)*
Normal	1.0 µL	–	–
Decreased	1.0 µL	20 µL	102 µL
Increased	1.0 µL	–	–

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

Calibration

Calibrators	S1: H_2O
	S2: C.f.a.s. PAC
Calibration mode	Linear
Calibration frequency	Automatic full calibration - after reagent lot change
	Full calibration - as required following quality control procedures

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

Traceability: This method has been standardized against an internal standard material.

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. 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 IU/mL.

Limitations - interference

Criterion: Recovery within ±10 % of initial value at an antistreptolysin O activity of 200 IU/mL.

Icterus:[15] 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:[15] No significant interference up to an H index of 1000 (approximate hemoglobin concentration: 621 µmol/L or 1000 mg/dL).

Lipemia (Intralipid):[15] No significant interference up to an L index of 1000. 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 180 IU/mL.

High-dose hook effect: No false result occurs up to an antistreptolysin O activity of 4000 IU/mL.

Drugs: No interference was found at therapeutic concentrations using common drug panels.[16,17]

In very rare cases, gammopathy, in particular type IgM (Waldenström’s macroglobulinemia), may cause unreliable results.[18]

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 further instructions, refer to the operator’s manual.

Limits and ranges

Measuring range

20-600 IU/mL

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:6.1 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 6.1.

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Lower limits of measurement

Limit of Blank, Limit of Detection and Limit of Quantitation

Limit of Blank	= 20 IU/mL
Limit of Detection	= 20 IU/mL
Limit of Quantitation	= 20 IU/mL

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 activity 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 activity antisptreptolysin O samples.

Expected values[19]

Adults	up to 200 IU/mL
Children	up to 150 IU/mL

In some cases of streptococcal infections, particularly skin infections, there may be no observable increase in the ASO titer. As antistreptolysin O is only detectable in 85 % of all patients with rheumatic fever, the determination of anti-streptococcal deoxyribonuclease antibodies and anti-streptococcal hyaluronidase antibodies may also be necessary.[19]

An appropriate evaluation of streptococcal infection is possible only if the test is repeated after 1 or 2 weeks.[20] Both clinical and laboratory findings should be correlated in reaching a diagnosis.

ASO levels are age dependent and change with geographic location and with the local frequency of streptococcal infections.[21,22]

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 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.

Repeatability	*Mean* *IU/mL*	*SD* *IU/mL*	*CV* *%*
PCCC1[a)]	117	2.41	2.1
PCCC2[b)]	251	2.44	1.0
Human serum 1	47.0	1.56	3.3
Human serum 2	86.7	3.49	4.0
Human serum 3	190	2.50	1.3
Human serum 4	307	3.37	1.1
Human serum 5	527	4.91	0.9

Intermediate precision	*Mean* *IU/mL*	*SD* *IU/mL*	*CV* *%*
PCCC1[a)]	121	2.87	2.4
PCCC2[b)]	248	3.59	1.5
Human serum 1	47.0	1.84	3.9
Human serum 2	86.7	3.75	4.3
Human serum 3	190	3.21	1.7
Human serum 4	307	4.29	1.4
Human serum 5	527	6.96	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) and **cobas c** 703 analyzer(s).

Method comparison

Antistreptolysin O values for human serum 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).

Sample size (n) = 68

Passing/Bablok[23]	Linear regression
y = 1.047x - 9.04 IU/mL	y = 1.055x - 11.8 IU/mL
τ = 0.981	r = 0.999

The sample concentrations were between 21 and 545 IU/mL.

Antistreptolysin O values for human serum 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).

Sample size (n) = 65

Passing/Bablok[23]	Linear regression
y = 1.021x + 0.0368 IU/mL	y = 1.018x - 1.08 IU/mL
τ = 0.980	r = 0.999

The sample concentrations were between 21.9 and 586 IU/mL.

Antistreptolysin O values for human serum samples obtained on a **cobas c** 703 analyzer (y) were compared with those determined using the corresponding reagent on a **cobas c** 503 analyzer (x).

Sample size (n) = 75

Passing/Bablok[23]	Linear regression
y = 0.976x - 1.50 IU/mL	y = 0.979x - 1.66 IU/mL
τ = 0.987	r = 0.999

The sample concentrations were between 36.4 and 593 IU/mL.

References

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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.

The Summary of Safety & Performance Report can be found here: https://ec.europa.eu/tools/eudamed

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see navifyportal.roche.com for definition of symbols used):

CONTENT	Contents of kit
→	Volume for reconstitution
GTIN	Global Trade Item Number
Rx only	For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

CE 0123

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