SARS-CoV-2 Total Antibody

Gentian SARS-CoV-2 Total Antibody Reagent Kit

REF 1801

For *in vitro* diagnostic use by laboratory professionals.

This document describes the general use of the product above. For instrument specific settings, please refer to the application notes available upon request from marketing@gentian.com.

Intended Purpose

The Gentian SARS-CoV-2 Total Antibody Immunoassay is a turbidimetric assay intended for the in vitro quantitative detection of total antibodies to the SARS-CoV-2 spike protein (S1-subunit) in human plasma and serum samples. The Gentian SARS-CoV-2 Total Antibody Immunoassay is intended for use on automated clinical analysers by laboratory professional users.

The Gentian SARS-CoV-2 Total Antibody Immunoassay is intended as an aid in identifying patients with an adaptive immune response to SARS-CoV-2, indicating recent or prior infection, or a successful response to vaccination. The Gentian SARS-CoV-2 Total Antibody Immunoassay should not be used to diagnose acute SARS-CoV-2 infection.

Summary and Explanation of Test

SARS-CoV-2 (severe acute respiratory syndrome coronavirus 2) causes COVID-19 (coronavirus disease 19) and has after its first case in 2019 spread in a world-wide pandemic and infected million causing the deaths of hundred thousand of people.[1] SARS-CoV-2 is an RNA virus of the corona-virus family belong to the same subfamily as seasonal coronaviruses causing the common cold. SARS-CoV-2 has 4 main structural proteins are nucleocapsid protein (N), envelope protein (E), membrane protein (M), and the major surface protein, spike protein (S), which contains the receptor-binding domain (RBD) that binds to the cellular receptors, like ACE2, during the infection.[2]

Molecular testing to detect the virus and serological testing to detect SARS-CoV-2 specific antibodies has been and will continue to be important in disease management[1]. SARS-CoV-2 antibodies levels rise within 2 weeks in most patients, where IgA and IgM rise earlier than IgG, but IgG levels are more stable overtime.[1,3-5] Also, the IgG levels are often higher in more severe cases than in mild cases.[6]

Serology testing can be a valuable tool to estimate the level of protection in each individual and the society.[7] Serological studies will be important to further increase the knowledge about vaccination response, immunity in the community and correlation of antibody levels and immunity.[8] Serological testing will be an important tool for public health measures including community surveillance, vaccination periodization and vaccination follow up.[7-9]

Calibrator Standardisation

The Gentian SARS-CoV-2 Total Antibody Calibrator is standardised against the WHO International Standard for anti-SARS-CoV-2 IgG (NIBSC code 20/136).

Assay Principle

The Gentian SARS-CoV-2 Total Antibody Immunoassay is a particle-enhanced turbidimetric immunoassay (PETIA). The lithium heparin plasma or serum sample is mixed with SARS-CoV-2 Total Antibody immunoparticles. Anti-S1 antibodies from the sample and the SARS-CoV-2 S1 protein from the immunoparticles solution bind to form aggregates that increase the turbidity of the solution. The degree of turbidity is proportional to the concentration of Anti-S1 antibodies, which can be quantified via an established standard calibration curve.

Assay Kit Components

Products provided	
Gentian SARS-CoV-2 Total Antibody Reagent Kit • R1 Assay Buffer (25 mL) • R2 Immunoparticles (4.5 mL)	REF 1801
Products required, but not provided	
Gentian SARS-CoV-2 Total Antibody Calibrator Kit (6 levels x 0.5 mL)	REF 1851
Gentian SARS-CoV-2 Total Antibody Control Kit (2 levels x 0.5 mL)	REF 1819

All products are ready for use.

Composition

Reaction Buffer 1 (R1, 25 mL inactive ingredient): SARS-CoV-2 Total Antibody assay buffer. R1 is a MOPS [3-(N-Morpholino)-propane sulfonic acid] buffered saline and preserved with ProClin® 950.

Reaction Buffer 2 (R2, 4.5 mL active ingredient): SARS-CoV-2 Total Antibody immunoparticles. R2 contains SARS-CoV-2 S1 recombinant antigen, which is covalently attached to polystyrene nanoparticles. The solution is preserved with ProClin® 950.

Hazards identification

Hazard pictograms (CLP): GHS07

Signal word (CLP): Warning
Contains: 2-methylisothiazol-3(2H)-one

Hazard statements (CLP):
H317 - May cause an allergic skin reaction.
Precautionary statements (CLP):
P280 - Wear eye protection, protective gloves, protective clothing.
P302+P352 - IF ON SKIN: Wash with plenty of soap and water.
P305+P351+P338 - IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P337+P313 - If eye irritation persists: Get medical advice/attention.
P333+P313 - If skin irritation or rash occurs: Get medical advice/attention.
P362+P364 - Take off contaminated clothing and wash it before reuse.

To obtain the SDS (safety data sheet), please contact Gentian at marketing@gentian.com.

Warnings and Precautions

1. Contains substances from human or animal origin and should be considered as potentially infectious material. Handle with caution and discard following local regulations.
2. Reagents containing MOPS/Tween (R1) can be irritating to eyes, respiratory tract and skin. Handle with due caution and do not ingest.

Additional Handling Instructions

1. This test is for *in vitro* use only and must be handled by qualified personnel.
2. Use only validated and approved instrument applications.
3. Do not use products after the expiration date has passed.
4. Do not mix reagents of different lots or interchange caps of reagents, controls, calibrators, and lots.
5. Tighten caps carefully back on after use of reagents, calibrators, and controls to avoid evaporation.

Reagent Storage and Stability

All products provided for the Gentian SARS-CoV-2 Total Antibody Immunoassay must be stored at 2-8°C. The expiry date is printed on the labels. Using an BS240 instrument (Mindray), the on-board stability of the Gentian SARS-CoV-2 Total Antibody Reagents was found to be at least 8 weeks in a study using a protocol based on the CLSI guideline EP25.

Specimen Collection and Handling

Recommended sample material is lithium heparin and EDTA plasma or serum. It is recommended to analyse the samples as fresh as possible. Mix samples well before analysing.

Performance Characteristics

The results refer to the main validation of the Gentian SARS-CoV-2 Total Antibody Immunoassay on a BS240/BS40 Pro instrument (Mindray) at one instrument site with one lot of reagents, unless otherwise stated. For the instrument specific performance characteristics, please refer to the instrument specific application notes.

Measuring Range

The measuring range of the Gentian SARS-CoV-2 Total Antibody Immunoassay is approximately 50 - 1000 BAU/mL, with a security zone of up to 3000 BAU/mL. The exact measuring range is calibrator- and instrument specific, please refer to the analytical value sheet for the lot specific calibrator values and the instrument specific application notes.

Analytical Sensitivity

The analysis of the analytical sensitivity of the Gentian SARS-CoV-2 Total Antibody Immunoassay was tested in a study including 6 samples run in 10 replicates over 3 days using a protocol based on the CLSI guideline EP17. The limit of quantification (LoQ) is defined as the lowest concentration of an analyte that can be reliably detected and at which the total error meets the requirements for accuracy. The LoQ was measured as 38 BAU/mL in lithium-heparin plasma and 26 BAU/mL in serum samples. For the instrument specific LoQ, please refer to the instrument specific application notes.

Precision

The total precision of the Gentian SARS-CoV-2 Total Antibody Immunoassay was measured with a CV of <10% for samples >165 BAU/mL in a study using a protocol based on the CLSI guideline EP05. For the instrument specific total precision, please refer to the instrument specific application notes.

Analytical Specificity and Limitations

No interference is detected for this product with haemoglobin, intralipid, bilirubin, HAS, lactoferrin, calcium, biotin heparin or disodium EDTA at the tested concentrations in a study using a protocol based on the CLSI guideline EP07. For the instrument specific interference, please refer to the instrument specific application notes. No cross reactivity was with Hepatitis B surface antigen (HbsAg), Hepatitis C (HCV) antibody, Anti-Influenza B and Anti-Adenovirus, Anti-Influenza A, Human Coronavirus 229E, OC43, NL63, and HKU1, Rheumatoid Factor (RF) IgG, Anti-nuclearantibodies (ANA), CMV IgM and HIV in a study including 13 samples using a protocol based on the CLSI guideline EP07.

Assay Procedure

Application Notes

Applications of the Gentian SARS-CoV-2 Total Antibody Immunoassay have been established on several clinical chemistry analysers. Detailed, validated application notes describing the procedures for installation and analysis on specific instruments are available upon request from marketing@gentian.com. For instructions on how to install a new application, consult the instrument manual. Maintenance, operation and precautions must be handled in accordance with the specific instrument manual.

Reagent Preparation

The reagents are ready for use. Mix the reagents gently before placing them into the assigned reagent positions. The reagent bottles may fit directly into the instrument unless otherwise stated in the application notes.

Calibration Curve Establishment

The calibration curve can either be created from a dilution series that is automatically prepared by the instrument or by using a 6-point pre-diluted calibrator kit as specified in the instrument manual. Calibrator values are lot specific, and calibration must be performed whenever a new lot is used. The values assigned to the calibrators are provided in the analytical value sheet. A new calibration should be performed according to the instrument specific calibration curve stability or when a new reagent lot is used.

QC Controls

The Gentian SARS-CoV-2 Total Antibody Controls should be assayed every day that the test is in use to validate the calibration curve. The controls have lot specific concentration ranges that must be met before measuring samples. The assigned value ranges are given in the analytical value sheet. If the control values measured are not valid, repeat the control measurements. Recalibrate if necessary. If the calibration cannot be performed without error, or valid control values cannot be reproduced, contact the local distributor for support.

Measuring Patient Samples

When a valid calibration curve has been established and the control values are within the valid range, the lithium heparin and EDTA plasma or serum sample may be measured. Ensure that the minimum sample volume is present in the sample cups/tubes and assay the samples according to the instructions given in the instrument manual.

Results

The results are calculated automatically by the instrument for all applications established for the Gentian SARS-CoV-2 Total Antibody Immunoassay. The results are presented in BAU/mL.

Clinical Performance

Sensitivity

A total of 414 samples from vaccinated or infected donors was analysed on the BS240 Pro instrument (Mindray) of which 8 returned false negative results returning a sensitivity of 98.0%.

Sensitivity = 100 – ((False negative/True positive)*100)

	No. of samples	No. <10 BAU/mL	Sensitivity	95% CI
Positive vaccinated	234	7	96.9%	[94.0%, 98.5%]
Positive infected	180	1	99.4%	[96.9%, 99.9%]
Total	**414**	**8**	**98.0%**	**[96.2%, 99.0%]**

Specificity

A total 621 samples from pre-pandemic donors, including samples from hospitalised and healthy donors as well as potential interfering samples and cross-reacting samples from pregnant women, donors with HCV and ANA antibodies and infected with Influenza A and B, RSV or endemic human corona virus types (antibodies positive samples against hCoV-229E, -OC43, -NL63 and -HKU1, were analysed on the BS240 Pro instrument (Mindray) of which 6 returned false positive results returning a specificity of 99.0%.

Specificity = 100 – ((False positive/True negative)*100)

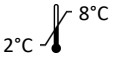
	No. of samples	No. >50 BAU/mL	Specificity	95% CI
Pre-pandemic hospitalised	298	2	99.3%	[97.6%, 99.8%]
Pre-pandemic healthy	234	4	98.3%	[95.7%, 99.3%]
Interfering samples	38	0	100%	[90.8%, 100.0%]
Cross reacting samples	51	0	100%	[93.0%, 100.0%]
Total	**621**	**6**	**99.0%**	**[97.9%, 99.6%]**

Reference Range

< 10 BAU/mL: negative
> 50 BAU/mL: positive

A negative result indicates the absence of S1-specific SARS-CoV-2 antibodies or levels below the detection limit of this test. A negative result does not completely exclude a previous infection and could occur during an acute infection prior to seroconversion or due to decrease of antibodies over time. A positive result indicates the presence of S1-specific SARS-CoV-2 antibodies due to exposure to SARS-CoV-2 or an immune reaction after a vaccination. In the grey zone between 10 and 50 BAU/mL the result is inconclusive. The results should always be assessed in conjunction with the patient's medical history, clinical examination and medical judgment.

Symbols Key

	Temperature Limit
	Use by Date
	Consult Instructions for Use
	Manufacturer
	CE Mark
IVD	*In Vitro* Diagnostic Medical Device
LOT	Lot Number
REF	Catalogue Number
UDI	Unique Device Identifier
CONTENTS	Contents
R1	R1 Assay Buffer
R2	R2 Immunoparticles
	Warning

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References

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3. Bohn, M. K., T. P. Loh, et al. Clinical Chemistry and Laboratory Medicine (CCLM) 58(12): 2001-2008.
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5. Ma, H., W. Zeng, et al. Cell Mol Immunol 17(7): 773-775.
6. Zhao, J., Q. Yuan, H. et al. Clin Infect Dis 71(16): 2027-2034.
7. Bonanni, P., R. Cantón, D. et al. COVID 1(1): 20-38.
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Serious Incidents

Please notify your manufacturer and competent authority if any serious incidents have occurred in relation to the device.

Modification from the Previous Version

First version

Date of issue

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