RightSign®

SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette Package Insert

REF ISNB-C41	English

A rapid test for the qualitative detection of neutralizing antibodies to SARS-CoV-2 in whole blood, serum, or plasma.
For professional in vitro diagnostic use only.

【INTENDED USE】

The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette is a rapid chromatographic immunoassay for the qualitative detection of neutralizing antibodies to SARS-CoV-2 in human whole blood, serum, or plasma as an aid in the diagnosis of the presence of neutralizing antibodies to SARS-CoV-2.

【SUMMARY】

The novel coronaviruses belong to the β genus. COVID-19 is an acute respiratory infectious disease. People are generally susceptible. Currently, the patients infected by the novel coronavirus are the main source of infection; asymptomatic infected people can also be an infectious source. Based on the current epidemiological investigation, the incubation period is 1 to 14 days, mostly 3 to 7 days. The main manifestations include fever, fatigue and dry cough. Nasal congestion, runny nose, sore throat, myalgia and diarrhea are found in a few cases.
The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette is a rapid test that utilizes a combination of Spike protein antigen coated colored particles for the detection of neutralizing antibodies to SARS-CoV-2 in human whole blood, serum or plasma.

【PRINCIPLE】

The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette is a qualitative membrane based immunoassay for the detection of neutralizing antibodies to SARS-CoV-2 in whole blood, serum or plasma. In this test procedure, neutralizing antibodies capture reagent is immobilized in the test line region of the test. After specimen is added to the specimen well of the strip, it reacts with Spike protein antigen coated particles in the test. This mixture migrates chromatographically along the length of the test and interacts with the immobilized neutralizing antibodies capture reagent. If the specimen contains neutralizing antibodies to SARS-CoV-2, a colored line will appear in the test line region, indicating a positive result. If the specimen does not contain neutralizing antibodies to SARS-CoV-2, a colored line will not appear in this region, indicating a negative result. To serve as a procedural control, a colored line will always appear in the control line region, indicating that proper volume of specimen has been added and membrane wicking has occurred.

【REAGENTS】

The test cassette contains to specific Spike protein antigen conjugated gold colloid particles and neutralizing antibodies capture reagent coated on the membrane.

【PRECAUTIONS】

- For professional in vitro diagnostic use only. Do not use after expiration date.
- Do not eat, drink or smoke in the area where the specimens or kits are handled.
- Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout the procedure and follow the standard procedures for proper disposal of specimens.
- Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
- The used tests, specimens and potentially contaminated material should be discarded according to the local regulations.
- Humidity and temperature can adversely affect results.

【STORAGE AND STABILITY】

The kit can be stored at room temperature or refrigerated (2-30°C). The test cassette is stable through the expiration date printed on the sealed pouch. The test cassette must remain in the sealed pouch until use. DO NOT FREEZE. Do not use beyond the expiration date.

【SPECIMEN COLLECTION AND PREPARATION】

- The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette can be performed using whole blood, serum, or plasma.
- Whole blood or plasma could be collected with tube containing Heparin or Citrate.
- To collect Fingerstick Whole Blood Specimens:
- Wash the patient's hand with soap and warm water or clean with an alcohol swab. Allow to dry.
- Massage the hand without touching the puncture site by rubbing down the hand towards the fingertip of the middle or ring finger.
- Puncture the skin with a sterile lancet. Wipe away the first sign of blood.
- Gently rub the hand from wrist to palm to finger to form a rounded drop of blood over the puncture site.
- Add the Fingerstick Whole Blood specimen to the test cassette by using a dropper, capillary or micropipette measuring 10ul. The dropper provided with the test dispenses approximately 10ul in one drop even if more blood is aspirated in the dropper or capillary.
- Separate serum or plasma from blood as soon as possible to avoid hemolysis. Use only clear, non-hemolyzed specimens.
- Testing should be performed immediately after specimen collection. Do not leave the specimens at room temperature for prolonged periods. Serum and plasma specimens may be stored at 2-8°C for up to 3 days. For long-term storage, specimens should be kept below -20°C. Whole blood collected by venipuncture should be stored at 2-8°C if the test is to be run within 2 days of collection. Do not freeze whole blood specimens. Whole blood collected by fingerstick should be tested immediately.
- Bring specimens to room temperature prior to testing. Frozen specimens must be completely thawed and mixed well prior to testing. Specimens should not be frozen and thawed repeatedly.
- If specimens are to be shipped, they should be packed in compliance with federal regulations for transportation of etiologic agents.

【MATERIALS】

Materials provided

Test cassettes	Droppers or capillaries
Buffer	Package insert

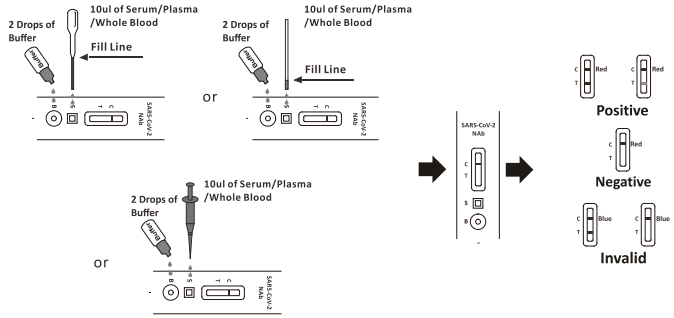
Materials required but not provided

Specimen collection containers	Centrifuge (for plasma only)
Micropipette	Timer
Lancets (for fingerstick whole blood only)	

【DIRECTIONS FOR USE】

Allow the test cassette, specimen, buffer, and/or controls to reach room temperature (15-30°C) prior to testing.

1. Bring the pouch to room temperature before opening. Remove the test cassette from the sealed pouch and use it within one hour.
2. Place the test cassette on a clean and level surface.
   - For Serum or Plasma or Whole Blood Specimens:
     - To use a dropper/ capillary: Hold the dropper/ capillary vertically, draw the specimen up to the Fill Line (approximately 10μl), and transfer the specimen to the specimen well (S) of the test cassette, then add 2 drops of buffer (approximately 80μl) to the buffer well (B) and start the timer. Avoid trapping air bubbles in the specimen well.
     - To use a micropipette: Pipette and dispense 10μl of specimen to the specimen well (S) of the test cassette, then add 2 drops of buffer (approximately 80μl) to the buffer well (B) and start the timer.
3. Wait for the colored line(s) to appear. The test result should be read at 10 minutes. Do not interpret the result after 20 minutes.



【INTERPRETATION OF RESULTS】

(Please refer to the illustration above)

POSITIVE:* **Two lines appear**. The colored line in the control line region (C) changes from **Blue to Red**, and other colored lines should appear in test line region (T).
*NOTE: The intensity of the color in the test line region will vary depending on the concentration of neutralizing antibodies to SARS-CoV-2 in the specimen. Therefore, any shade of color in the test line region should be considered positive.
NEGATIVE: The colored line in the control line region (C) changes from **Blue to Red**. No line appears in test line region (T).
INVALID: Control line (C) is still completely or partially blue, and fails to completely change from **Blue to Red**. Insufficient buffer volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the procedure with a new test cassette. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

【QUALITY CONTROL】

An internal procedural control is included in the test. A colored line appearing in the control line region (C) is an internal valid procedural control, it confirming adequate membrane wicking. Control standards are not supplied with this kit; however, it is recommended that positive and negative controls be tested as a good laboratory practice to confirm the test procedure and to verify proper test performance.

【LIMITATIONS】

1. The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole Blood/Serum/Plasma) is for in vitro diagnostic use only. The test should be used for the detection of neutralizing antibodies to SARS-CoV-2 in whole blood, serum or plasma specimens only.
2. The best time to detect the presence of neutralizing antibodies is 10 days from the second vaccination day (COVID-19 Vaccination are given on two days usually with a gap of 28 days – Here the number of days to be calculated from the second vaccination day) in case of vaccination with vaccines containing inactivated viral particles and 15 days from the second vaccination day (COVID-19 Vaccination are given on two days usually with a gap of 28 days – Here the number of days to be calculated from the second vaccination day) in case of mRNA vaccines.
3. Results from the SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole Blood/Serum/Plasma) should not be used as the sole basis to diagnose or exclude the presence of neutralizing antibodies to SARS-CoV-2.
4. The continued presence or absence of neutralizing antibodies cannot be used to determine the success or failure of therapy or vaccination.
5. Results from immunosuppressed patients should be interpreted with caution.
6. As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.
7. This test should not be used for blood donor screening.
8. This test has not been reviewed by the FDA.

【PERFORMANCE CHARACTERISTICS】

Generally, the higher the titer of neutralizing antibody is, the better the individual protection is. Microneutralization Assay (MNA_{50}) is an established method of assessing the humoral response. Positive results obtained with 1:10 dilution in MNA_{50} assay indicates that the individuals are already protected and the same with 1:20 dilution indicates that the individuals have strong protection. Therefore, MNA_{50} assay at 1:10 dilution was selected as the comparison method to evaluate the efficacy of the SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole Blood/Serum/Plasma)

1st The cutoff of MNA_{50} is maintained at 1:10 dilution and the clinical characteristics are showed below:

In order to evaluate the clinical performance of SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole Blood/Serum/Plasma), the comparator Microneutralization Assay (MNA) and SARS-CoV-2 Neutralizing Antibody Elisa Kit were used. The cutoff for the MNA comparator tests was established as indicated below:

Value Result (Dilution titer)	Result	Test Result Interpretation
⩾1:10	Positive	SARS-CoV-2 Neutralizing antibodies are detected at 50% viral neutralization.
＜1:10	Negative	SARS-CoV-2 Neutralizing antibodies are not detected at 50% viral neutralization.

Part 1: Clinical Performance using MNA_{50} titer as the comparator method with samples from convalescent patients, or healthy unvaccinated individuals

A total of 48 samples were retrospectively collected from convalescent patients, or healthy unvaccinated individuals (30 MNA_{50} positive and 18 MNA_{50} negative) and were evaluated with the SARS-CoV-2 Neutralizing Antibody rapid test cassette.

Item		Microneutralization Assay (MNA_{50})		Total Result
SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole blood/Serum/Plasma)	Result	Positive	Negative	
	Positive	30	0	30
	Negative	0	18	18
Total Result		30	18	48

Relative Sensitivity: 30/ (0+30) =100.0% (95%*CI: 90.5%~100.0%);
Relative Specificity: 18/ (18+0) =100.0% (95%*CI: 84.7%~100.0%);
Accuracy: (30+18)/ (30+0+0+18) =100.0% (95%*CI: 93.9%~100.0%);
*CI means confidence interval.

Part 2: Clinical Performance using MNA_{50} titer as the comparator method with samples from vaccinated individuals, or healthy unvaccinated individual

A total of 47 samples were collected from vaccinated individuals (Inactivated SARS-CoV-2 Vaccine), or healthy unvaccinated individuals (29 MNA_{50} positive and 18 MNA_{50} negative) and were evaluated with the SARS-CoV-2 Neutralizing Antibody rapid test cassette.

Item		Microneutralization Assay (MNA_{50})		Total Result
SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole blood/Serum/Plasma)	Result	Positive	Negative	
	Positive	29	0	29
	Negative	0	18	18
Total Result		29	18	47

Relative Sensitivity: 29/ (0+29) =100.0% (95%*CI: 90.2%~100.0%);
Relative Specificity: 18/ (18+0) =100.0% (95%*CI: 84.7%~100.0%);
Accuracy: (29+18)/ (29+0+0+18) =100.0% (95%*CI: 93.8%~100.0%);
*CI means confidence interval.

2nd The cutoff of MNA_{50} is maintained at 1:20 (dilution titer) and the clinical characteristics are showed below:

In order to evaluate the clinical performance of SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole Blood/Serum/Plasma), the comparator Microneutralization Assay (MNA) and SARS-CoV-2 Neutralizing Antibody Elisa Kit were used. The cutoff for the MNA comparator tests was established as indicated below:

Value Result (Dilution titer)	Result	Test Result Interpretation
⩾1:20	Positive	SARS-CoV-2 Neutralizing antibodies are detected at 50% viral neutralization.
＜1:20	Negative	SARS-CoV-2 Neutralizing antibodies are not detected at 50% viral neutralization.

Part 1: Clinical Performance using MNA_{50} titer as the comparator method with samples from convalescent patients, or healthy unvaccinated individuals

A total of 48 samples were retrospectively collected from convalescent patients, or healthy unvaccinated individuals (30 MNA_{50} positive and 18 MNA_{50} negative) and were evaluated with the SARS-CoV-2 Neutralizing Antibody rapid test cassette.

Item		Microneutralization Assay (MNA_{50})		Total Result
	Result	Positive	Negative	
SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole blood/Serum/Plasma)	Positive	30	0	30
	Negative	0	18	18
Total Result		30	18	48

Relative Sensitivity: 30/ (0+30) =100.0% (95%*CI: 90.5%~100.0%);
Relative Specificity: 18/ (18+0) =100.0% (95%*CI: 84.7%~100.0%);
Accuracy: (30+18)/ (30+0+0+18) =100.0% (95%*CI: 93.9%~100.0%);
*CI means confidence interval.

Part 2: Clinical Performance using MNA_{50} titer as the comparator method with samples from vaccinated individuals, or healthy unvaccinated individual

A total of 47 samples were collected from vaccinated individuals (Inactivated SARS-CoV-2 Vaccine), or healthy unvaccinated individuals (26 MNA_{50} positive and 21 MNA_{50} negative) and were evaluated with the SARS-CoV-2 Neutralizing Antibody rapid test cassette.

Item		Microneutralization Assay (MNA_{50})		Total Result
	Result	Positive	Negative	
SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole blood/Serum/Plasma)	Positive	26	3	29
	Negative	0	18	18
Total Result		26	21	47

Relative Sensitivity: 26/ (0+26) =100.0% (95%*CI: 89.1%~100.0%);
Relative Specificity: 18/ (18+3) =85.7% (95%*CI: 63.7%~97.0%);
Accuracy: (26+18)/ (26+3+0+18) =93.6% (95%*CI: 82.5%~98.7%);
*CI means confidence interval.

3rd Clinical Performance using Elisa kit as the comparator method with samples from vaccinated individuals, or healthy unvaccinated individual

A total of 60 samples were collected from vaccinated individuals(Inactivated SARS-CoV-2 Vaccine), or healthy unvaccinated individuals (30 Elisa positive and 30 Elisa negative) and were evaluated with the SARS-CoV-2 Neutralizing Antibody rapid test cassette.

Item		Elisa Kit		Total Result
	Result	Positive	Negative	
SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (Whole blood/Serum/Plasma)	Positive	30	0	30
	Negative	0	30	30
Total Result		30	30	60

Relative Sensitivity: 30/ (0+30) =100.0% (95%*CI: 90.5%~100.0%);
Relative Specificity: 30/ (30+0) =100.0% (95%*CI: 96.3%~100.0%);
Accuracy: (30+30)/ (30+0+0+30) =100.0% (95%*CI: 95.1%~100.0%);
*CI means confidence interval.

Cross-reactivity

The SARS-CoV-2 Neutralizing Antibody Rapid Test Cassette (whole blood/Serum/Plasma) has been tested for anti-influenza A virus, anti-influenza B virus, anti-RSV, anti-Adenovirus, HBsAb, anti-Syphilis, anti-H. Pylori, anti-HIV, anti-HCV and HAMA positive specimens. The results showed no cross-reactivity.

Interfering Substances

The following potentially interfering substances were added to SARS-CoV-2 neutralizing antibody negative and spiked positive specimens.

Analytes	Concentration	Result	
		Negative Specimen	Spiked with Positive Specimen
Acetaminophen	20 mg/dL	Negative	Positive
Caffeine	20 mg/dL	Negative	Positive
Albumin	2 g/dL	Negative	Positive
Acetylsalicylic Acid	20 mg/dL	Negative	Positive
Gentisic Acid	20 mg/dL	Negative	Positive
Ethanol	1%	Negative	Positive
Ascorbic Acid	2g/dL	Negative	Positive
Creatine	200mg/dl	Negative	Positive
Bilirubin	1g/dL	Negative	Positive
Hemoglobin	1000mg/dl	Negative	Positive
Oxalic Acid	60mg/dL	Negative	Positive
Uric acid	20mg/ml	Negative	Positive

None of the substances at the concentration tested interfered in the assay.

【BIBLIOGRAPHY】

1. Weiss SR, Leibowitz JL. Coronavirus pathogenesis. Adv Virus Res 2011; 81:85-164.
2. Cui J, Li F, Shi ZL. Origin and evolution of pathogenic coronaviruses. Nat Rev Microbiol 2019; 17:181-192.
3. Su S, Wong G, Shi W, et al. Epidemiology, genetic recombination, and pathogenesis of coronaviruses. TrendsMicrobiol 2016;24:490-502.

Index of Symbols

	Consult Instruction for use		Tests per kit	EC REP	Authorized Representative
IVD	For in vitro diagnostic use only		Use by		Do not reuse
2°C 30°C	Store between 2-30°C	LOT	Lot Number	REF	Catalog #
	Do not use if package is damaged				

EC REP
CMC Medical Devices & Drugs S.L.
C/ Horacio Lengo N° 18, CP 29006,
Málaga, Spain.

Hangzhou Biotest Biotech Co.,Ltd.
No.17, Futai Road, Zhongtai Street,
Yuhang District, Hangzhou -311121
P.R. China

Number: RP5369701
Effective date: 2021-02-19