TRUSTline COVID-19 Ab Rapid Test - Cassette

REF AR0185C IVD

for qualitative detection of anti-SARS-CoV-2 antibodies in human serum and plasma or whole blood

INTENDED USE

The TRUSTline COVID-19 Ab Rapid Test is a single use lateral flow immunoassay rapid test intended for qualitative detection of anti-SARS-CoV-2 antibodies in human serum and plasma or whole blood containing EDTA, heparin or citrate anti-coagulants. The TRUSTline COVID-19 Ab Rapid Test is intended for use as an aid in identifying individuals with an adaptive immune response to SARS-CoV-2 infection. At this time, it is unknown for how long antibodies persist following infection and if the presence of antibodies confers protective immunity .

Results are for the detection of SARS CoV-2 antibodies. Antibodies to SARS-CoV-2 are generally detectable in blood several days after initial infection, although the duration of time that antibodies are present post-infection is not well characterized. Individuals may have detectable virus present for several weeks following seroconversion.

SUMMARY AND EXPLANATION OF THE TEST

SARS-CoV-2 belongs to the broad family of coronaviruses which are capable of causing illnesses ranging from the common cold to more severe diseases[1]. SARS-CoV-2 infections cause COVID-19 disease. The infected patients have a wide range of clinical symptoms, from little to no symptoms, to fever, tiredness and dry cough, possibly leading to severe sickness and death. Most patients recover without special treatment. Around 1 out of every 6 patients who get COVID-19 become seriously ill and develop difficulty breathing. Older people and those with underlying medical problems, like high blood pressure, heart problems or diabetes, are more likely to develop serious illness.

Human-to-human transmission of the virus has been confirmed and occurs primarily via respiratory droplets from coughs and sneezes within a range of about 6 feet (1.8 m). Viral RNA has also been found in stool samples from patients. It's possible that the virus can be infectious even during the incubation period, but this has not been proven[2].

Currently, the laboratory method for detecting SARS-CoV-2 infection is RT-PCR. However, this method requires sophisticated equipment and highly trained laboratory technicians. Moreover, viral load decreases rapidly 9 or 10 days after onset of symptoms. During the acute phase of infection, the titer of IgM to SARS-CoV-2 rises rapidly and peaks around 2-3 weeks after the infection. SARS-CoV-2 specific IgG antibodies appear shortly after IgM and persist for months[3]. It is unknown if SARS-CoV-2 infection leads to lifetime immunity or if a 2nd infection is possible. Nevertheless, the SARS- CoV-2 specific antibodies are useful markers for immune response and epidemiologic survey.

The TRUSTline COVID-19 Ab Rapid Test detects anti-SARS-CoV-2 antibodies in human serum, plasma or whole blood. The test can be performed within 15 minutes by minimally skilled personnel without the use of cumbersome laboratory equipment.

TEST PRINCIPLE

The TRUSTline COVID-19 Ab Rapid Test is a lateral flow chromatographic immunoassay. The test strip in the cassette consists of: 1) a colored conjugate pad containing SARS-CoV-2 recombinant antigens conjugated with colloidal gold (SARS-CoV-2 conjugates) and a control antibody conjugated with colloidal gold, 2) a nitrocellulose membrane strip containing a test line (Ab line) and a control line (C line). The Ab line is pre-coated with both SARS- CoV-2 antigens and anti-human antibodies for capturing anti-SARS-CoV-2 antibodies, and the C line is pre-coated with a control line antibody.

When an adequate volume of specimen is dispensed into the sample well of the test cassette, the specimen migrates by capillary action along the cassette strip. Anti-SARS-CoV-2 antibodies, if present in the specimen, will bind to the SARS-CoV-2 conjugates. The immunocomplex is then captured by the pre-coated reagents, forming a colored Ab line, indicating an anti-SARS-CoV-2 antibody positive test result. A positive result indicates an immune response to SARS-CoV-2, and suggests a late or previous COVID-19 infection.

Absence of the test line suggests a negative result. Each test contains an internal control (C line) which should exhibit a colored line of the control antibodies regardless of color development on any of the test lines. If the C line does not develop, the test result is invalid, and the specimen must be retested with another device.

REAGENTS AND MATERIALS PROVIDED

1. Individually sealed foil pouches containing:
   a. One cassette device
   b. One desiccant
2. Disposable capillary tubes, marked for 10 µL and 20 µL
3. Detection buffer (tris-based buffered solution with preservatives)
4. Instructions for Use

MATERIALS REQUIRED BUT NOT PROVIDED

1. Clock or watch or other timing device.
2. Pipettor capable of delivering 10-20 µL of sample, that can be used instead of the disposable capillary tube for greater accuracy.
3. Sterile lancets, sterile gauze and wipes for fingerstick whole blood specimens.
4. Collection devices for venous whole blood, serum, plasma.
5. Disposable gloves, biohazard disposal container.

WARNINGS AND PRECAUTIONS

1. Read these Instructions for Use completely before performing the test. Failure to follow the instructions could lead to inaccurate test results.
2. Do not open the sealed pouch, unless ready to conduct the assay.
3. Once the pouch is opened, it should be used within 30 minutes to avoid possible failure caused by the absorption of moisture.
4. Do not use expired devices or components.
5. Do not use the components of any other type of test kit as a substitute for the components in this kit.
6. Do not use hemolyzed blood specimens for testing.
7. Use only one specimen per device. Do not combine specimens.
8. Wear protective clothing and disposable gloves while handling the kit reagents and clinical specimens. Wash hands thoroughly after testing.
9. Follow the US CDC Universal Precautions for prevention of transmission of HIV, HBV and other bloodborne pathogens.
10. Do not smoke, drink, or eat in areas where specimens or kit reagents are being handled.
11. Dispose of all specimens and materials used to perform the test as bio-hazardous waste.
12. Handle external controls in the same manner as patient specimens.
13. Read test results 10-15 minutes after a specimen is applied to the sample well of the device. Reading the test result after 15 minutes should be considered invalid and must be repeated.
14. Do not perform the test in a room with strong air flow, i.e. an electric fan or strong air-conditioning.

REAGENT PREPARATION AND STORAGE INSTRUCTIONS

All reagents are ready to use as supplied. Store unused test devices unopened at 1-30°C. If stored at 2-8°C, ensure that the test device is brought to room temperature before opening. The test device is stable until the expiration date printed on the sealed pouch. Do not freeze the kit or expose the kit to temperatures above 30°C.

SPECIMEN COLLECTION AND HANDLING

Consider any materials of human origin as potentially infectious, and handle them with standard biosafety procedures.

Plasma/Serum

Step 1: Collect venous blood by venipuncture into collection tubes containing EDTA, citrate or heparin anticoagulants for plasma, or collection tubes containing no anticoagulants for serum.

Step 2: A) To prepare plasma specimens, centrifuge the blood and carefully withdraw the plasma into a new pre-labeled tube.
B) To prepare serum specimens, allow blood to clot, centrifuge and carefully withdraw the serum into a new pre-labeled tube.

Test serum/plasma specimens as soon as possible after collection, or store refrigerated at 2-8°C for up to 3 days or frozen at -20°C for longer storage.

Avoid multiple freeze-thaw cycles. Prior to testing, bring frozen specimens to room temperature slowly and mix gently. Specimens containing visible particulate matter should be clarified by centrifugation before testing.

Do not use specimens demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid possible interference with result interpretation.

Whole Blood

Step 1: Whole blood can be obtained by either fingertip puncture using a safety lancet, or by venipuncture. Collect venous blood into a collection tube containing EDTA, citrate or heparin anticoagulants. Do not use hemolyzed blood for testing.

Test whole blood specimens as soon as possible after collection, or store refrigerated (2-8°C). Specimen must be tested within 24 hours of collection.

Note: Do not test specimens demonstrating gross lipemia, gross hemolysis or turbidity in order to avoid possible interference with the assay result.

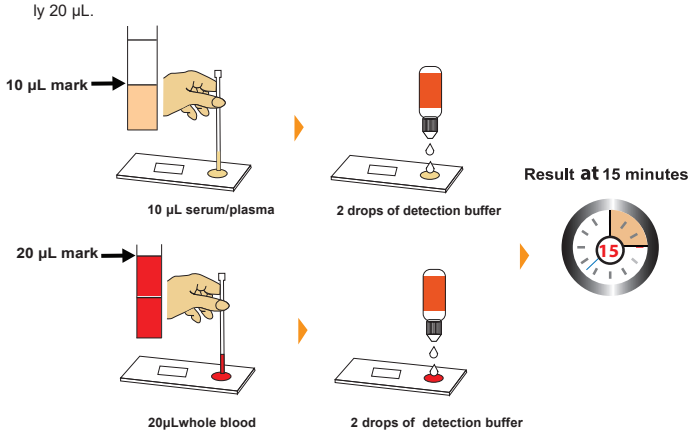
ASSAY PROCEDURE

Step 1: Ensure that specimen and test components are equilibrated to room temperature before testing. If frozen, mix the specimen well after thawing, prior to performing the assay.

Step 2: When ready to test, open the pouch at the notch and remove device. Place the test device on a clean, flat surface.

Step 3: Label the device with the specimen's ID number.

Step 4: **The volume of specimen used in the test is different when using whole blood versus serum or plasma.**
For serum/plasma: Fill the capillary tube with serum or plasma up to, but not exceeding, the mark (10 µL mark) as shown in the image below. The volume of specimen is approximately 10 µL.
For whole blood: Fill the capillary tube with whole blood up to, but not exceeding, the mark (20 µL mark) as shown in the image below. The volume of specimen is approximately 20 µL.



Step 5: Holding the capillary tube vertically, dispense the entire amount of specimen into the center of the sample well, ensuring that there are no air bubbles. **For better precision, specimen can be transferred using a pipette capable of delivering a volume of 10 µL for serum or plasma, or 20 µL for whole blood specimens.**

Step 6: Immediately add 2 drops (approximately 70-100 µL) of detection buffer into the sample well of the test cassette, ensuring that there are no air bubbles.

Step 7: Set up timer.

Step 8: Read results at 15 minutes. Positive results may be visible within 2 minutes. ***All results must be confirmed at 15 minutes, outside the 15 minutes window should be considered invalid and must be repeated. Discard used device after interpreting the results following local laws governing the disposal of devices.***

QUALITY CONTROL

1. An internal procedural control is included in the test. A colored line appearing in the C line is an internal procedural control. It confirms sufficient sample volume, adequate membrane wicking and correct procedural technique.
2. External positive and negative controls are not supplied with this kit; however, external positive and negative controls should be tested consistent with good laboratory practice to confirm the test procedure and to verify proper test performance.

INTERPRETATION OF ASSAY RESULT

1. **NEGATIVE RESULT:** If only the C line is present, the absence of the test line (Ab) indicates that no SARS-CoV-2 antibodies are detected. The result is negative or non-reactive.

2. **POSITIVE RESULT:** In addition to the presence of the C line, if the Ab line develops, the test indicates the presence of SARS-CoV-2 antibodies. The result is positive or reactive.

3. **INVALID:** If no C line develops, the assay is invalid regardless of any color in the test line (Ab) as indicated below. Repeat the assay with a new device.

PERFORMANCE CHARACTERISTICS

1. In-house evaluation

Clinical performance was evaluated with 13 different positive specimens. TRUSTline COVID-19 Ab Rapid Test showed positive reactivity (100% sensitivity) to all the specimens. Specificity of the TRUSTline COVID-19 Ab Rapid Test was evaluated by testing the test device with 500 random negative clinical specimens collected from healthy individuals. 100 % specificity was obtained for TRUSTline COVID-19 Ab Rapid Test.

2. External evaluation

Clinical performance was evaluated with 100 different positive specimens and 100 different negative specimens. TRUSTline COVID-19 Ab Rapid Test showed 92% sensitivity and 100% specificity to all the specimens.

	TRUSTline COVID-19 Ab Rapid Test		
CMIA & RT PCR	**Positive**	**Negative**	**Total**
Positive	92	8	100
Negative	0	100	100
Total	92	108	200

LIMITATIONS OF TEST

1. The TRUSTline COVID-19 Ab Rapid Test is limited to the qualitative detection of anti-SARS-CoV-2 virus antibodies in human serum, plasma and whole blood. The intensity of the test band does not have linear correlation with the antibody titer in the specimen.
2. Results from antibody testing should not be used to diagnose or exclude acute SARS-CoV-2 infection or inform infection status.
3. Reactive results must be confirmed with another available method and interpreted in conjugation with patient's clinical manifestation
4. Negative results do not preclude acute SARS CoV-2 infection. If acute infection is suspected direct testing for SARS CoV-2 is necessary.
5. The Assay Procedure and the Test Result Interpretation must be followed closely when testing the presence of antibodies to SARS-CoV-2 virus in serum, plasma and whole blood. Failure to follow the procedure may lead to inaccurate results.
6. The performance of the test has been validated using the specimen volumes corresponding to the respective marks on the capillary tube. Exceeding the mark when loading the specimen could lead to false positive results.
7. Unusually high titer of heterophile antibodies or rheumatoid factor present in some specimens may affect the expected results[4,5]. Factors, such as operational error can also potentially induce false results.
8. Positive results may be due to past or present infection with non-SARS-CoV-2 coronavirus strains, such as coronavirus HKU1, NL63, OC43, or 229E.

REFERENCES

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Index of Symbols

Consult instructions for use	REF Catalogue number	Use-by date
IVD For *in vitro* diagnostic use only	LOT Batch code	Tests per kit
Temperature limit 1-30 °C	Do not re-use	Keep dry
Manufacturer	Date of manufacture	
Do not use if package is damaged	Keep away from sunlight	

ATHENESE-Dx
Early diagnosis for better life

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PI-AR0185C Rev. B
Effective date: 01.10.2020
English version