COVID-19 IgG/IgM RAPID TEST (SERUM / PLASMA / WHOLE BLOOD)

Instructions for Use

10108883
BARCODE FOR RTR USE ONLY

REF R0180C

2°C 30°C

IVD

INTENDED USE

The *Aria* COVID-19 IgG/IgM Rapid Test is a single use lateral flow immunoassay rapid test intended for qualitative detection and differentiation of anti-SARS-CoV-2 IgG and IgM antibodies in human serum and plasma or whole blood containing EDTA, heparin or citrate anti-coagulants. The *Aria* COVID-19 IgG/IgM Rapid Test is intended for use as an aid in identifying individuals with an adaptive immune response to SARS-CoV-2, indicating recent or prior 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. IgG and IgM 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.

The sensitivity of *Aria* COVID-19 IgG/IgM Rapid Test early after infection is unknown. Negative results do not preclude acute SARS-CoV-2 infection. If acute infection is suspected, direct testing for SARS-CoV-2 is necessary.

False positive results for *Aria* COVID-19 IgG/IgM Rapid Test may occur due to cross-reactivity from pre-existing antibodies or other possible causes. Due to the risk of false positive results, confirmation of positive results should be considered using second, different assays such as ELISA or Chemiluminescent IgG/IgM Test.

For prescription use only. For *in vitro* diagnostic use only.

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 *Aria* COVID-19 IgG/IgM Rapid Test detects anti-SARS-CoV-2 IgG and IgM 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 *Aria* COVID-19 IgG/IgM 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 two test lines (G and M lines) and a control line (C line). The G line is pre-coated with antibodies for the detection of anti-SARS-CoV-2 IgG, the M line is pre-coated with antibodies for the detection of anti-SARS-CoV-2 IgM, 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 IgG, if present in the specimen, will bind to the SARS-CoV-2 conjugates. The immunocomplex is then captured by the pre-coated anti-human IgG, forming a colored G line, indicating an anti-SARS-CoV-2 IgG positive test result, suggesting a recent infection or a past infection. Anti-SARS-CoV-2 IgM, if present in the specimen, will bind to the SARS-CoV-2 conjugates. The immunocomplex is then captured by the pre-coated anti-human IgM, forming a colored M line, indicating an anti-SARS-CoV-2 IgM positive test result and suggesting an acute SARS-CoV-2 infection. An IgM and IgG double positive result suggests a late acute infection.

Absence of any of the test lines (G or M) 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, 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

For *In Vitro* Diagnostic Use

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 performing the test.
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 2-30°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 specimens as soon as possible after collection. If not tested immediately, serum and plasma specimens can be stored 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 or by venipuncture. Collect venous blood into a collection tube containing EDTA, citrate or heparin anticoagulants. Do not use hemolyzed blood for testing.

Test specimens as soon as possible after collection. If not tested immediately, whole blood specimens should be stored refrigerated (2-8°C), and 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.

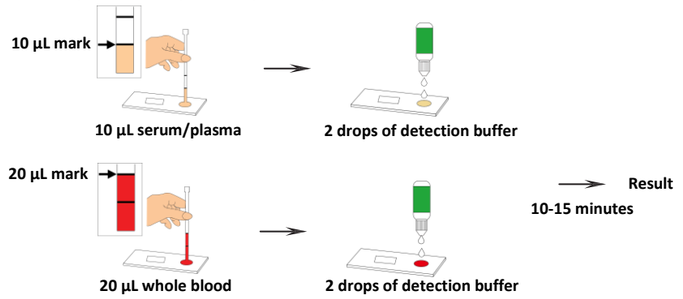
For whole blood: Fill the capillary tube with whole blood up to, but not exceeding, the mark (**20 µL mark**) as shown in the image below. The volume of specimen is approximately 20 µL.

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 10-15 minutes. Positive results may be visible within 2 minutes. All results must be confirmed at 15 minutes. ***Any results interpreted outside the 10-15 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.

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INTERPRETATION OF ASSAY RESULT

1. **NEGATIVE RESULT:** If only the C line is present, the absence of any color in both test lines (M and G) indicates that there is no SARS-CoV-2 IgG or IgM antibodies detected. The result is negative or non-reactive.

2. **POSITIVE RESULT:** In addition to the presence of the C line, if the G or M line develops, or both G and M lines develop, the test indicates the presence of SARS-CoV-2 IgG and/or IgM antibody. The result is positive or reactive. Borderline results producing a faint band that cannot be interpreted as positive must be repeated to confirm the presence of the visible test line bands indicating positive results.

Positive results should be confirmed with alternative testing method(s) and clinical findings for a diagnosis decision.

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

PERFORMANCE CHARACTERISTICS

1. Clinical Performance

1.1. Positive Agreement

Endemic, subjects symptomatic or with suspected Infection

A total of 210 specimens (serum or plasma) were collected during the COVID-19 pandemic from subjects either symptomatic or with a suspected infection. All subjects were confirmed COVID-19 positive by real time PCR (RT-PCR). Positive agreement for all subjects is shown in the table below:

Days after Hospital Admission	Number of samples	COVID-19 RT-PCR Result	*Aria* COVID-19 IgG/IgM Rapid Test Result		
			Positive for IgG	Positive for IgM	Positive for IgG or IgM
1-7	7	Positive	86% (6/7)	100% (7/7)	100% (7/7)
8-14	4	Positive	75% (3/4)	75% (3/4)	75% (3/4)
15-21	10	Positive	80% (8/10)	80% (8/10)	80% (8/10)
22-28	30	Positive	93% (28/30)	93% (28/30)	93% (28/30)
29-35	52	Positive	98% (51/52)	92% (48/52)	98% (51/52)
36-42	61	Positive	100% (61/61)	95% (58/61)	100% (61/61)
43-49	37	Positive	100% (37/37)	89% (33/37)	100% (37/37)
50-56	9	Positive	100% (9/9)	78% (7/9)	100% (9/9)
Total	210	Positive	**96.7% (203/210)** (95% CI: 93.3%-98.4%)	**91.4% (192/210)** (95% CI: 86.9%-94.5%)	**97.1% (204/210)** (95% CI: 93.9%-98.7%)

1.2. Negative Agreement

Endemic, subjects symptomatic or with suspected Infection

A total of 270 specimens (serum or plasma) were collected during the COVID-19 pandemic from subjects either symptomatic or with a suspected infection. All subjects were confirmed COVID-19 negative by real time PCR. Negative agreement for all subjects is shown in the table below:

Number of samples	COVID-19 RT-PCR Result	*Aria* COVID-19 IgG/IgM Rapid Test Result		
		Negative for IgG	Negative for IgM	Negative for IgG and IgM
270	Negative	**98.1% (265/270)** (95% CI: 95.7%-99.2%)	**99.3% (268/270)** (95% CI: 97.3%-99.8%)	**97.8% (264/270)** (95% CI: 95.2%-99.0%)

2. Cross-reactivity

No false positive test results for either anti-SARS-CoV-2 virus IgG and IgM were observed on 5-12 specimens from the following disease states or specific conditions:

HBV	HCV	HIV	HBsAg	Tuberculosis	Zika	Dengue	Influenza A	ANA
EBV	VZV	CMV	Syphilis	Chikungunya	Measles	Mumps	Influenza B	

3. Class Specificity

Recombinant human anti-SARS-CoV-2 IgG and IgM antibodies were serially-diluted in COVID-19 negative plasma and tested on the *Aria* COVID-19 IgG/IgM Rapid Test. No cross-reactivity between COVID-19 IgG and IgM was observed, as shown in the following table:

Sample	Dilution	IgG	IgM
Negative control	Undiluted	Negative	Negative
Anti-SARS-CoV-2 **IgG**	1:2	Positive	Negative
	1:10	Positive	Negative
	1:50	Positive	Negative
	1:250	Positive	Negative
	1:1250	Negative	Negative
Anti-SARS-CoV-2 **IgM**	1:2.5	Negative	Positive
	1:10	Negative	Positive
	1:40	Negative	Positive
	1:160	Negative	Positive
	1:640	Negative	Negative

The results from the class specificity study above also suggest no hook effect in the *Aria* COVID-19 IgG/IgM Rapid Test, as no negative impact was observed when testing samples with increased concentration of either IgG or IgM antibodies.

4. Matrix equivalency

The performance of the *Aria* COVID-19 IgG/IgM Rapid Test in different specimen matrices was evaluated in samples from 13 convalescent confirmed positive COVID-19 and from 14 COVID-19 negative subjects. Capillary whole blood was collected by fingerstick puncture and 5 different venous matrices where collected by venipuncture: venous whole blood, serum, and 3 plasma matrices (K2-EDTA, sodium citrate and lithium heparin). The concordance observed was 100% for all matrices, for both positive and negative specimen. The results are shown in the following table:

Matrix		Number of positive results on *Aria* COVID-19 IgG/IgM Rapid Test			
		Positive samples		Negative samples	
		IgG	IgM	IgG	IgM
Capillary whole blood		100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)
Venous whole blood		100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)
Serum		100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)
Plasma	K2-EDTA	100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)
	Sodium citrate	100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)
	Lithium heparin	100% (13/13)	100% (13/13)	0% (0/14)	0% (0/14)

5. Interference

No interference was observed with the potentially interfering substances listed below at the indicated concentration:

Substance	Concentration	Substance	Concentration
Hemoglobin	10 mg/mL	Caffeine	58 µg/mL
Bilirubin	0.4 mg/mL	Aspirin	60 mg/dL
Triglycerides	15 mg/mL	Biotin	200 ng/dL
Sodium heparin	125 U/mL	Ethanol	4 mg/mL
Sodium citrate	3.8%	Sodium EDTA	3.4 µM
Human Serum Albumin	60 mg/mL		

LIMITATIONS OF TEST

1. The *Aria* COVID-19 IgG/IgM Rapid Test is limited to the qualitative detection of anti-SARS-CoV-2 virus IgG and IgM 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. The *Aria* COVID-19 IgG/IgM Rapid Test is intended for use as an aid in identifying individuals with an adaptive immune response to SARS-CoV-2, indicating recent or prior infection. The test should not be used to diagnose the COVID-19 disease.
3. 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 from individual subjects. Failure to follow the procedure may lead to inaccurate results.
4. The performance of *Aria* COVID-19 IgG/IgM Rapid Test has not been validated in patients who have received vaccination or have been treated with antibody drug to SARS-CoV-2 coronavirus.
5. 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.
6. 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.
7. Negative results do not rule out SARS-CoV-2 infection, particularly in those who have been in contact with the virus. Follow-up testing with a molecular diagnostic should be considered to rule out infection in these individuals.
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.
9. Results from antibody testing should not be used as the sole basis to diagnose or exclude SARS-CoV-2 infection or to inform infection status.
10. Not for the screening of donated blood.

REFERENCES

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4. Hansen, H. J., Sharkey, R. M., Sullivan, C. L., & Goldenberg, D. M. (1993). HAMA interference with murine monoclonal antibody-based immunoassays. JOURNAL OF CLINICAL IMMUNOASSAY, 16(4), 294-299.
5. Levinson, S. (1992). The nature of heterophilic antibodies and their role in immunoassay interference. J. Clin. Immunoassay, 15, 108-115.

Index of Symbols

	Consult instructions for use	IVD	For *in vitro* diagnostic use only		Use by
REF	Catalog #	LOT	Lot Number	N	Tests per kit
2°C 30°C	Store between 2-30°C	EC REP	Authorized Representative		Do not reuse
	Manufacturer		Date of manufacture		

CTK Biotech, Inc.
13855 Stowe Drive
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E-mail: info@ctkbiotech.com

EC REP **MDSS GmbH**
Schiffgraben 41
30175 Hannover, Germany

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Date released: 2020-05-13
English version

For Export Only, Not For Re-sale in the USA.