Monkeypox Virus Antigen Rapid Test
Package Insert

REF IMXG-402 | **English**

CE IVD

For professional in vitro diagnostic use only.

【INTENDED USE】

The Monkeypox Virus Antigen Rapid Test is a rapid chromatographic immunoassay intended for the qualitative detection of monkeypox virus A29L antigen in human serum, plasma, whole blood, rash exudate and throat swab specimens. It is intended for use as an aid in the diagnosis of monkeypox virus infection. Results are for the detection of monkeypox virus antigen. Positive results indicate the presence of monkeypox virus antigen.

【SUMMARY】

Monkeypox is considered a zoonotic disease with transmission primarily occurring from animals, such as rodents and primates, to humans. However, limited sustained human-to-human transmission has been observed with up to 6 generations of human-to-human transmission being previously identified. Transmission of the virus can occur through contact with bodily fluids, wounds on the skin or internal mucosal surfaces, respiratory droplets, or contaminated objects. Consumption of inadequately cooked meat or other products from infected animals may also pose increased risks of infection.[1]

The incubation period for monkeypox can range from 5–21 days but usually falls within 7–14 days. Clinical presentation of monkeypox can be similar to chickenpox, caused by varicella-zoster virus.[2] Symptoms usually begin within 5 days of infection with fever and chills, headache, muscle aches, back pain, fatigue, and swollen lymph nodes (lymphadenopathy), the latter symptom differentiating monkeypox from smallpox and chickenpox. About 1–3 days, sometimes longer, after the initial onset of symptoms, a rash or lesions can appear, usually beginning on the face and spreading throughout the body, often to the extremities rather than the trunk. Notably, monkeypox lesions can appear on the palms of the hands and soles of the feet (75% of cases). Most individuals with monkeypox experience rash with 1 to >100 skin lesions, but some do not experience these lesions.[1]

In most patients, symptoms of monkeypox are usually self-limiting and spontaneously resolve within 14-21 days. However, symptoms can be severe and require medical care.[1]

【PRINCIPLE】

The Monkeypox Virus Antigen Rapid Test is a qualitative, lateral flow immunoassay for the detection of monkeypox virus A29L antigen in human serum, plasma, whole blood, rash exudate and throat swab specimens. The membrane is pre-coated with anti-MPX A29L on the test line region. During testing, monkeypox virus antigen in the serum, plasma, whole blood, rash exudate and throat swab specimen reacts with the particle coated with MPX A29L antibody. The mixture migrates upward on the membrane chromatographically by capillary action to react with monkeypox virus antibody on the membrane and generate a colored line. The presence of this colored line in the test region indicates a positive result, while its absence indicates 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 contains MPX A29L antibody coated particles as a detection reagent and anti-MPX A29L coated with cellulose nitrate membrane as a capture reagent.

【PRECAUTIONS】

1. This package insert must be read completely before performing the test. Failure to follow directions in package insert may yield inaccurate test results.
2. For professional *in vitro* diagnostic use only. Do not use after the expiration date.
3. Do not eat, drink or smoke in the area where the specimens or kits are handled.
4. Do not use the test if the pouch is damaged.
5. Handle all specimens as if they contain infectious agents. Observe established precautions against microbiological hazards throughout the collection, handling, storage and disposal of patient samples and the disposal of used kit contents.
6. Wear protective clothing such as laboratory coats, disposable gloves and eye protection when specimens are assayed.
7. Wash hands thoroughly after testing.
8. Please ensure that appropriate amounts of samples are used for testing. Too much or too little may lead to deviation of results.
9. The used test should be discarded according to local regulations.
10. Humidity and temperature can adversely affect results.
11. Where use of a centrifuge is required for a procedure, safety cups or sealed rotors should be used.

【STORAGE AND STABILITY】

- Store as packaged in the sealed pouch at room temperature or refrigerated (2-30°C).
- The test is stable until the expiration date printed on the sealed pouch.
- The test must remain in the sealed pouch until use.
- **DO NOT FREEZE.**
- Do not use beyond the expiration date.

【SPECIMEN COLLECTION AND PREPARATION】

Rash Exudate Swab or Throat Swab

For rash exudate, wipe the rash exudate 5 laps with a sterile swab. For throat swab, insert swab into the posterior pharynx and tonsillar areas. Rub swab over both tonsillar pillarsand posterior oropharynx 5 laps and avoid touching the tongue, teeth, and gums. Swab specimens should be tested as soon as possible after collection.

If swabs are not been processed immediately, it is highly recommended the swab sample is placed into a dry, sterile and tightly sealed plastic tube for storage. The swab specimen in dry and sterile condition is stable for up to 24 hours at 2-8°C.

Whole Blood, Serum or Plasma

Collect whole blood specimen into a collection tube (with specified anticoagulant, namely EDTA K2, heparin sodium, sodium citrate or potassium oxalate) according to standard venous blood sampling process.

Separate the serum or plasma from blood as soon as possible to avoid hemolysis. Only clear, non-hemolyzed specimens can be used.

- Testing should be performed immediately after the specimens have been collected. 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 10 days. For long term storage, they should be kept below -20°C. Whole blood specimens should be stored at 2-8°C if the test is to be run within 3 days after collection. Do not freeze whole blood specimens.
- 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 local regulations covering the transportation of etiologic agents.
- EDTA-K2, Heparin sodium, Citrate sodium and Potassium Oxalate can be used as the anticoagulant for collecting the specimen.

【MATERIALS】

Material Provided

• Test Cassettes	• Buffer	• Package Insert
• Sterile Swabs	• Workstation	

Materials Required but Not Provided

• Centrifuge	• Timer	• Pipette
• Alcohol Pad	• Lancet	• Droppers

【DIRECTIONS FOR USE】

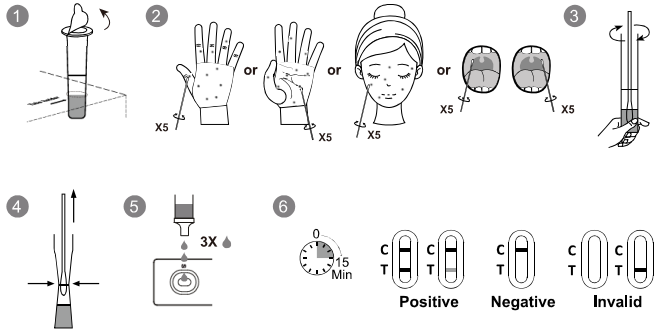
Allow the test, specimen and buffer equilibrate to room temperature (15-30°C) prior to testing.

1. Remove the test cassette from the foil pouch and use it as soon as possible. Best results will be obtained if the assay is performed within one hour.
2. Place the test on a flat and clean surface, and run test as below for different specimen.

Rash Exudate Swab or Throat Swab

For **Rash Exudate** or **Throat Swab** specimen:

1. Remove the cover of the tube with extraction buffer and place the tube in the workstation.
2. For rash exudate, wipe the rash exudate 5 laps with a sterile swab. For throat swab, insert swab into the posterior pharynx and tonsillar areas. Rub swab over both tonsillar pillarsand posterior oropharynx 5 laps and avoid touching the tongue, teeth, and gums.
3. Place the swab into the extraction tube. Rotate the swab for **10-15 seconds** while pressing the head against the inside of the tube to release the antigen in the swab.
4. Remove the swab while squeezing the swab head against the inside of the extraction tube as you remove it to expel as much liquid as possible from the swab. Discard the swab in accordance with your biohazard waste disposal protocol.
5. Fit the tube tip or close the cap onto the tube, then invert the extraction tube and add **3 drops of specimen** (approximately 75 µL) into the specimen well (S) and then start the timer.
6. Wait for the colored line (s) to appear. **Read results at 15 minutes**. Do not interpret the result after 20 minutes.

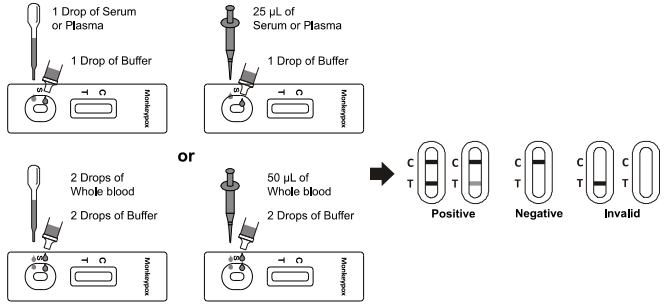


Whole Blood, Serum or Plasma

1. For **Serum or Plasma** specimen:
   - Use a dropper: Hold the dropper vertically, transfer **1 full drops** (approximately 25 µL) of **Serum or Plasma** to the Specimen well (S). Then add **1 drop of buffer** (approximately 25 µL) to the Specimen well (S), and start the timer.
   - Use a pipette: Transfer **25 µL of Serum or Plasma** to the Specimen well(S), then add **1 drop of buffer** (approximately 25 µL) to the Specimen well (S), and start the timer.

   For **Whole blood** specimen:
   - Use a dropper: Hold the dropper vertically, transfer **2 full drops** (approximately 50 µL) of **Whole blood** to the Specimen well (S). Then add **2 drops of buffer** (approximately 50 µL) to the Specimen well (S), and start the timer.
   - Use a pipette: Transfer **50 µL of Whole blood** to the Specimen well(S), then add **2 drops of buffer** (approximately 50 µL) to the Specimen well (S), and start the timer.
2. Wait for the colored line(s) to appear. **Read results at 15 minutes**. Do not interpret the result after 20 minutes.

Note: It is suggested not to use the buffer beyond 6 months after opening the vial.



【INTERPRETATION OF RESULTS】

POSITIVE:* Two colored lines appear. One colored line should be in the control region (C) and another colored line should be in the test region (T). Positive result in the test region indicates monkeypox virus antigen was detected in the specimen.

***NOTE**: The intensity of the color in the test line region (T) will vary based on the amount of monkeypox virus antigen present in the sample. So any shade of color in the test region (T) should be considered positive.

NEGATIVE: One colored line appears in the control region (C). No colored line appears in the test line region (T).

INVALID: Control line fails to appear. Insufficient specimen volume or incorrect procedural techniques are the most likely reasons for control line failure. Review the procedure and repeat the test with a new test. If the problem persists, discontinue using the test kit immediately and contact your local distributor.

【QUALITY CONTROL】

Internal procedural controls are included in the test. A colored line appearing in the control region (C) is an internal procedural control. It confirms sufficient specimen volume and correct procedural technique. 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 DIRECTIONS FOR USE and the INTERPRETATION OF RESULTS must be followed closely when testing for the presence of monkeypox virus antigen in the serum, plasma, whole blood, rash exudate and throat swab specimen from individual subjects. For optimal test performance, proper sample collection is critical. Failure to follow the procedure may give inaccurate results.
2. The Monkeypox Virus Antigen Rapid Test is for *in vitro* diagnostic use only. This test should be used for detection of monkeypox virus antigen in serum, plasma, whole blood, rash exudate and throat swab specimens. Neither the quantitative value nor the rate of increase in the concentration of monkeypox virus antigen can be determined by this qualitative test.
3. The Monkeypox Virus Antigen Rapid Test will only indicate the presence of monkeypox virus antigen.
4. The hematocrit level of the whole blood can affect the test results. Hematocrit level needs to be between 25% and 65% for accurate results.
5. The test will show negative results under the following conditions: The titer of the monkeypox virus antigen in the sample is lower than the minimum detection limit of the test, or the monkeypox virus antigen has not appeared at the time of sample collection. It is recommended to re-sample the patient a few days later and test again.
6. The continued presence or absence of antigen cannot be used to determine the success or failure of a certain therapy.

【PERFORMANCE CHARACTERISTICS】

Sensitivity and Specificity

The Monkeypox Virus Antigen Rapid Test was compared with Monkeypox Virus PCR kit. The results were tabulated as below:

Method		PCR		Total Results
Monkeypox Virus Antigen Rapid Test	Results	Positive	Negative	
	Positive	15	1	16
	Negative	1	199	200
Total Results		16	200	216

Relative Sensitivity: 93.75% (95%CI*: 69.77%-99.84%) *Confidence Interval
Relative Specificity: 99.50% (95%CI*: 97.25%-99.99%)
Accuracy: 99.07% (95%CI*: 96.70%-99.89%)

Limitation of Detection

The Monkeypox Virus Antigen Rapid Test can detect out Monkeypox virus A29L antigen as low as 0.1 ng/mL.

Precision

Intra-Assay

Within-run precision has been determined by using 10 replicates of four specimens: a negative, a low positive, a middle positive and a high positive. The negative, low positive, middle positive and high positive were correctly identified >99% of the time.

Inter-Assay

Between-run precision has been determined by 10 independent assays on the same four specimens: a negative, a low positive, a middle positive and a high positive. Three different lots of the Monkeypox Virus Antigen Rapid Test have been tested over a 3-days period using negative, low positive, middle positive and high positive specimens. The specimens were correctly identified >99% of the time.

Cross-reactivity

The Monkeypox Virus Antigen Rapid Test has been tested for HAMA, Rheumatoid factor (RF), anti-HAV IgG, anti-Syphilis TP, anti-HIV, anti-*H. Pylori,* anti-Toxoplasma IgG, anti-Toxoplasma IgM, anti-Rubella IgG, anti-Rubella IgM, anti-CMV IgG, anti-CMV IgM, HBsAg, AFP, CEA, anti-Measles, anti-SARS-CoV-2 IgG positive specimens. The results showed no cross-reactivity.

The following culture virus strains were indicated no cross-reactivity with Monkeypox Virus Antigen Rapid Test.

No.	Culture Virus Strains	Concentration
1	Cowpox Virus	1E+05$TCID_{50}$/mL
2	Ectromelia Virus	1E+05$TCID_{50}$/mL
3	Varicella-zoster Virus	1E+03$TCID_{50}$/mL
4	Herpes Simplex Virus	1E+04$TCID_{50}$/mL
5	Vaccinia Virus	1E+05$TCID_{50}$/mL

Interfering Substances

The following compounds have been tested using the Monkeypox Virus Antigen Rapid Test and no interference was observed.

Triglyceride: 100 mg/dL Ascorbic Acid: 20 mg/dL Hemoglobin: 1000 mg/dL
Bilirubin: 60 mg/dL Total cholesterol: 15 mmol/L

【BIBLIOGRAPHY】

1. World Health Organization (WHO). Monkeypox Fact Sheet. Geneva: WHO; 2019. Accessed 18 May 2022. https://www.who.int/news-room/fact-sheets/detail/monkeypox
2. US Centers for Disease Control and Prevention (CDC). Monkeypox Signs and Symptoms. Updated July 16, 2021. Accessed May 18, 2022. https://www.cdc.gov/poxvirus/monkeypox/symptoms.html

Index of Symbols

	Consult instructions for use or consult electronic instructions for use		Contains sufficient for <n> tests		Temperature limit
	*In vitro* diagnostic medical device		Batch code		Catalogue number
	Authorized representative in the European Community/European Union		Use-by date		Do not re-use
	Do not use if package is damaged and consult instructions for use		Manufacturer		Caution

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