Rapid Response™

Human Monkeypox Virus Antibody Rapid Test
(Whole Blood/Serum/Plasma)

REF HMPV-13C25 Product Insert

A rapid test for the qualitative detection of antibodies (IgG and IgM) to Monkeypox Virus in whole blood, serum, or plasma.

For professional *in vitro* diagnostic use only.

Intended Use

The Rapid Response™ Human Monkeypox Virus Antibody Rapid Test is a rapid chromatographic immunoassay for the qualitative detection of IgG and IgM antibodies to Monkeypox Virus in human whole blood, serum, or plasma as an aid in the diagnosis of Monkeypox Virus infections.

Summary

Human monkeypox (HMPX), caused by the monkeypox virus (MPXV) which is a double-stranded DNA virus, a member of the orthopoxvirus genus within the Poxviridae family. It is a viral zoonotic disease, meaning that it can spread from animals to humans. It can also spread between people.
The incubation period of monkeypox can range from 5 to 21 days. The febrile stage of illness usually lasts 1 to 3 days with symptoms including fever, intense headache, lymphadenopathy (swelling of the lymph nodes), back pain, myalgia (muscle ache), and an intense asthenia (lack of energy). The febrile stage is followed by the skin eruption stage, lasting for 2 to 4 weeks. Lesions evolve from macules (lesions with a flat base) to papules (raised firm painful lesions) to vesicles (filled with clear fluid) to pustules (filled with pus), followed by scabs or crusts.
The Rapid Response™ Human Monkeypox Virus Antibody Rapid Test is a rapid test that utilizes a combination of Monkeypox Virus antigen coated colored particles for the detection of IgG and IgM antibodies to Monkeypox Virus in human whole blood, serum, or plasma.

Principle

The Rapid Response™ Human Monkeypox Virus Antibody Rapid Test is a qualitative membrane-based immunoassay for the detection of human monkeypox virus antibodies in whole blood, serum, or plasma. This test consists of two components, an IgG component and an IgM component. In the IgG component, anti-human IgG is coated in IgG test line region. During testing, the specimen reacts with monkeypox virus antigen-coated particles in the test cassette. The mixture then migrates upward on the membrane chromatographically by capillary action and reacts with the anti-human IgG in IgG test line region. If the specimen contains IgG antibodies to monkeypox virus, a colored line will appear in IgG test line region. In the IgM component, anti-human IgM is coated in IgM test line region. During testing, the specimen reacts with anti-human IgM. IgM antibodies to monkeypox virus, if present in the specimen, reacts with the anti-human IgM and the monkeypox virus antigen-coated particles in the test cassette, and this complex is captured by the anti-human IgM, forming a colored line in IgM test line region.
Therefore, if the specimen contains IgG antibodies to monkeypox virus, a colored line will appear in IgG test line region. If the specimen contains IgM antibodies to monkeypox virus, a colored line will appear in IgM test line region. If the specimen does not contain antibodies to monkeypox virus, no colored line will appear in either of the test line regions, indicating a negative result. To serve as a procedural control, a colored line will always change from Blue to Red in the control line region, indicating that membrane wicking has occurred.

Reagents

The test cassette contains monkeypox virus antigen conjugated gold colloid particles and anti-human IgM, anti-human IgG 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.

Materials

Materials provided

- Test cassettes
- Buffer
- Droppers
- Product insert

Materials required but not provided

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

Storage and Stability

The kit can be stored at room temperature or refrigerated (35.6-86°F; 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.

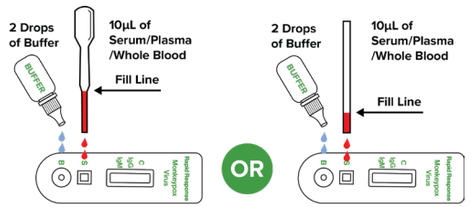
Collection and Storage of Specimens

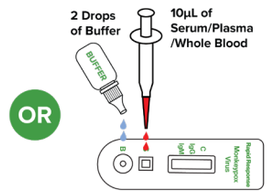
- The Rapid Response™ Human Monkeypox Virus Antibody Rapid Test can be performed using whole blood, serum, or plasma.
- **To collect Fingerstick Whole Blood Specimens:**
  - Wash the patient's hand with soap and warm water or clean with an alcohol swab. Allow to dry.
  - Massage the hand without touching 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 or micropipette measuring 10µl. The dropper provided with the test dispenses approximately 10µl in one drop even if more blood is aspirated in the dropper.
- 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 35.6-46.4°F (2-8°C) for up to 3 days. For long-term storage, specimens should be kept below -4°F (-20°C). Whole blood collected by venipuncture should be stored at 35.6-46.4°F (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.

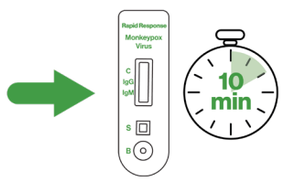
Test Procedure

Allow the test cassette, specimen, buffer, and/or controls to reach room temperature (59-86°F; 15-30°C) prior to testing.

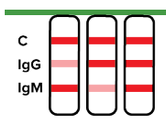
1. Bring the pouch to room temperature before opening. Remove the test cassette from the sealed pouch and use it within one hour.
2. Place the test cassette on a clean and level surface.
3. **For Serum/Plasma/Whole Blood Specimens:**
   - Hold the dropper vertically, draw the specimen up to the Fill 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.
4. 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.







Results Interpretation

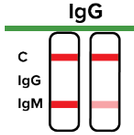


IgG and IGgM

IgG and IgM POSITIVE:* The colored line in the control line region (C) changes from Blue to Red, and two colored lines should appear in IgG test line region and IgM test line region. The color intensities of the lines do not have to match.

IgG

IgG POSITIVE:* The colored line in the control line region (C) changes from Blue to Red, and a colored line appears in IgG test line region. The result is positive for monkeypox virus specific-IgG.

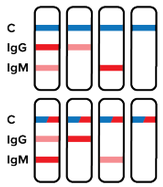


IgM

IgM POSITIVE:* The colored line in the control line region (C) changes from Blue to Red, and a colored line appears in IgM test line region. The result is positive for monkeypox virus specific-IgM antibodies.

***NOTE:** The intensity of the color in the IgG and/or IgM test line region(s) will vary depending on the concentration of monkeypox virus antibodies in the specimen. Therefore, any shade of color in the IgG and/or IgM test line region(s) should be considered positive.

NEGATIVE: The colored line in the control line region (C) changes from Blue to Red. No line appears in IgG and IgM test line region(s).



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, 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 Rapid Response™ Human Monkeypox Virus Antibody Rapid Test is for *in vitro* diagnostic use only. The test should be used for the detection of human monkeypox antibodies in whole blood, serum or plasma specimens only. Neither the quantitative value nor the rate of increase in monkeypox antibody concentration can be determined by this qualitative test.
2. In the early onset of fever, anti-monkeypox virus IgM concentrations may be below detectable levels.
3. The continued presence or absence of antibodies cannot be used to determine the success or failure of therapy.
4. Results from immunosuppressed patients should be interpreted with caution.
5. As with all diagnostic tests, all results must be interpreted together with other clinical information available to the physician.
6. Negative results do not rule out human monkeypox virus 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.
7. Positive results may be due to past or present infection with other Orthopoxvirus, such as cowpox virus, and smallpox virus.
8. Results from antibody testing should not be used as the sole basis to diagnose or exclude monkeypox virus infection or to inform infection status.
9. Not for the screening of donated blood.

Performance Characteristics

Sensitivity and Specificity

A clinical evaluation was conducted comparing the results obtained using the Rapid Response™ Human Monkeypox Virus Antibody Rapid Test to clinical performance. The study included 110 IgG specimens and 110 IgM specimens.

Item		Clinical performance		Total Result
Rapid Response™ Human Monkeypox Virus Antibody Rapid Test for IgG	Result	Positive	Negative	
	Positive	10	0	10
	Negative	0	100	100
Total Result		10	100	110

Relative Sensitivity: 10/10=100% (95%*CI: 74.1%~100.0%);
Relative Specificity: 100/100=100% (95%*CI: 97.0%~100.0%);
Accuracy: 110/110=100% (95%*CI: 97.3%~100.0%);
*CI means confidence interval.

Item		Clinical performance		Total Result
Rapid Response™ Human Monkeypox Virus Antibody Rapid Test for IgM	Result	Positive	Negative	
	Positive	10	0	10
	Negative	0	100	100
Total Result		10	100	110

Relative Sensitivity: 10/10=100% (95%*CI: 74.1%~100.0%);
Relative Specificity: 100/100=100% (95%*CI: 97.0%~100.0%);
Accuracy: 110/110=100% (95%*CI: 97.3%~100.0%);
*CI means confidence interval.

Cross-reactivity

The Rapid Response™ Human Monkeypox Virus Antibody Rapid Test has been tested for HBsAg, HBsAb, HBeAg, HBeAb, HBcAb, Anti-Syphilis, Anti-H. Pylori, Anti-HIV, Anti-HCV, Anti-TOXO, Anti-Rubella, Anti-CMV and Anti-HSV positive specimens. The results showed no cross-reactivity.

Interfering Substances

The following potentially interfering substances were added to Monkeypox Virus Antibody negative and positive specimens.

Acetaminophen	20 mg/dL
Acetylsalicylic Acid	20 mg/dL
Ascorbic Acid	2g/dL
Hemoglobin	1000mg/dl
Caffeine	20 mg/dL
Gentisic Acid	20 mg/dL
Creatine	200mg/dl
Oxalic Acid	60mg/dL
Albumin	2 g/dL
Ethanol	1%
Bilirubin	1g/dL
Uric acid	20mg/ml

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

Bibliography

1. World Health Organization (WHO). Laboratory testing for the monkeypox virus; 23 May 2022.
2. Harapan, H, Setiawan, A. M, et al. Confidence in managing human monkeypox cases in Asia: A cross-sectional survey among general practitioners in Indonesia. Acta Tropica, 206 (2020), 105450.
3. Ihekweazu, C, Yinka-Ogunleye, A, et al. Importance of epidemiological research of Monkeypox: is incidence increasing? Expert Review of Anti-infective Therapy (2020); 1478-7210.

Glossary of Symbols

Consult instructions for use
Test per Kit
Do Not Reuse
Store between 35.6°F to 86°F
Use by
REF Catalogue #
LOT Lot Number
IVD For in vitro diagnostic use only

BTNX Inc.
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