Mpox (Monkeypox)

Cue® Mpox (Monkeypox) Molecular Test
Instructions For Use
For Professional Use

Use with the Cue Health Monitoring System (Reader) and Cue Health App.

For use under an Emergency Use Authorization (EUA) only. For prescription use only. For in vitro diagnostic use.

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Instructions for Use

Summary and Explanation

Mpox is a disease caused by the monkeypox virus, which is a double-stranded DNA virus that belongs to the *Orthopoxvirus* genus in the family Poxviridae.[1] Mpox is spread to humans through close contact with an infected person or animal, or with material contaminated with the virus.[2] There are two distinct clades of mpox, clade I (Congo Basin – Central African Region) and clade II (West Africa).[1,2,3] The typical presentation of mpox consists of fever, rash and swollen lymph nodes.[2] Monkeypox virus was first isolated from a 9-month-old boy from the Democratic Republic of the Congo in 1970[4] and since then most mpox cases have been endemic in Central and West Africa.[3,5] Since early May 2022, confirmed cases (clade IIb) have been reported in countries that have not previously reported cases of mpox, and continue to be reported in several endemic countries.[6, 7,8]

The Cue Mpox (Monkeypox) Molecular Test is an in vitro diagnostic test that aids in the detection and diagnosis of monkeypox virus and is based on widely used isothermal nucleic acid amplification technology. The Cue Mpox (Monkeypox) Molecular Test contains primers and probes and internal controls used in molecular tests for the in vitro qualitative detection of monkeypox virus nucleic acid.

The Cue Mpox (Monkeypox) Molecular Test detects monkeypox nucleic acid from samples collected with the Cue Sample Wand directly from skin lesions or by dipping the Cue Sample Wand in previously collected direct skin lesion samples in viral transport media (VTM).

Intended Use

The Cue Mpox (Monkeypox) Molecular Test is an isothermal nucleic acid amplification assay for the qualitative detection of DNA from the monkeypox virus (clade I/II), in human lesion swab specimens (i.e., swabs of acute pustular or vesicular rash) from individuals suspected of mpox by their healthcare provider. The test is run using the Cue Health Monitoring System (Cue Reader), the Cue Mpox (Monkeypox) Molecular Test Cartridge, the Cue Sample Wand, and the Cue Health App on a compatible mobile smart device named on the Cue Health website at www.cuehealth.com.

Testing is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet requirements to perform high, moderate, or waived complexity tests. Testing is authorized for use at the Point of Care (POC), i.e., in patient care settings operating under a CLIA Certificate of Waiver, Certificate of Compliance, or Certificate of Accreditation.

Results are for the identification of monkeypox virus (clade I/II) DNA which is generally detectable in human pustular or vesicular lesion specimens during the acute phase of infection. Positive results are indicative of the presence of monkeypox virus (clade I/II) DNA; clinical correlation with patient history and other diagnostic information is necessary to determine patient infection status. Positive results do not rule out bacterial infection or co-infection with other viruses. The agent detected may not be the definite cause of disease. Negative results obtained with this device do not preclude monkeypox virus infection and should not be used as the sole basis for treatment or other patient management decisions. Negative results must be combined with clinical observations, patient history, and epidemiological information.

Laboratories within the United States and its territories are required to report test results to the appropriate public health authorities.

The Cue Mpox (Monkeypox) Molecular Test is intended for use by trained operators who are proficient in performing a test using the Cue Health Monitoring System.

The Cue Mpox (Monkeypox) Molecular Test is only for use under the Food and Drug Administration's Emergency Use Authorization.

Principles of the Procedure

The Cue Mpox (Monkeypox) Molecular Test utilizes isothermal nucleic acid amplification technology (NAAT) for the qualitative detection of monkeypox virus nucleic acid. This test detects monkeypox virus nucleic acid using a molecular amplification reaction that is an equivalent alternative amplification method to polymerase chain reaction (PCR). The Cue Mpox (Monkeypox) Molecular Test primers amplify a conserved region found twice (gene name OPG001) within the monkeypox virus genome enabling detection. The Cue Mpox (Monkeypox) Molecular Test forward primers are conjugated to biotin and the reverse primers are conjugated to Horseradish Peroxidase (HRP). RNase P serves as the internal control. The RNase P forward primer is conjugated to a small hapten, Digoxigenin (Dig). The RNase P reverse primer is conjugated to HRP.

The RNase P internal control has been designed to control for presence of human cellular material in the sample and proper assay execution including sample inhibition, amplification, and assay reagent function. If RNase P and monkeypox virus are not detected, the Cue Mpox (Monkeypox) Molecular Test will return an "Invalid" result. In positive samples where target amplification is strong, the internal control is ignored, and the target amplification serves as the control to confirm that the clinical sample was not inhibitory and that assay reagent performance was robust.

When the user inserts the Cue Sample Wand with a skin lesion sample into the Cue Test Cartridge, the test automatically begins. Heating, mixing, amplification, and detection take place within the cartridge. The current flow from the electrodes provides a semi-quantitative nanoampere measurement that is converted to a positive or negative result (based on a pre-determined cutoff). The Cue Mpox (Monkeypox) Molecular Test takes about 25 minutes from Sample Wand insertion to results.

Precautions - General

- This product is an *in vitro* diagnostic test, which means the test is performed outside the body.
- For use under emergency use authorization only.
- For prescription use only.
- This product has not been FDA cleared or approved but has been authorized by FDA under an EUA for use by authorized laboratories.
- This product has been authorized only for the detection of nucleic acid from monkeypox virus, not from any other viruses or pathogens.
- The emergency use of this product is only authorized for the duration of the declaration that circumstances exist justifying the authorization of emergency use of *in vitro* diagnostics for detection and/or diagnosis of infection with the monkeypox virus, including in vitro diagnostics that detect and/or diagnose infection with non-variola *Orthopoxvirus*, under Section 564(b)(1) of the Federal Food, Drug, and Cosmetic Act, 21 U.S.C. § 360bbb-3(b)(1), unless the declaration is terminated or authorization is revoked sooner.

- Laboratories within the United States and its territories are required to report all results to the appropriate public health authorities.
- The Cue Mpox (Monkeypox) Molecular Test Cartridge is used with the Cue Reader and Cue Health App.
- The Cue Reader needs to be on a level surface when the Cue Mpox (Monkeypox) Molecular Test Cartridge is inserted and while the test is running. Do not move the Cue Reader while the test is running.
- Only skin lesion swabs from papular (solid, raised with distinct borders), vesicular (fluid trapped under the epidermis), pustular (filled with yellow fluid) or macular (flat) lesions can be used with the Cue Mpox (Monkeypox) Molecular Test.
- Performance of the Cue Mpox (Monkeypox) Molecular Test has only been established for detection of monkeypox virus nucleic acid from samples collected with the Cue Sample Wand directly from skin lesions or by dipping the Cue Sample Wand in previously collected direct skin lesion samples in viral transport media (VTM). The performance of this test with other specimen types or samples has not been evaluated.
- Collection of lesion samples should be performed by healthcare professionals trained to collect lesion samples. Personal protective equipment (PPE) can help protect the skin and mucous membranes of the eyes, nose, and mouth should be worn by the healthcare professional collecting the lesion sample.
- Do not clean the lesion with ethanol or any other disinfectant prior to swabbing.
- Avoid procedures that could generate infectious aerosols.
- Do not use the Cue Mpox (Monkeypox) Molecular Test Cartridge past the Use By date on the Cartridge foil pouch label.
- Do not use the Cue Sample Wand past the Use By date on the Wand label.
- The Cue Health Monitoring System (Cartridge Reader) must be cleaned and disinfected after each use. See the Cue Health Monitoring System User Manual for instructions.
- Do not remove the Wand from the Cue Mpox (Monkeypox) Molecular Test Cartridge during testing or after the test has been performed.

Precautions - Cue Mpox (Monkeypox) Molecular Test Cartridge and Cue Sample Wand Handling

- Open the Cue Mpox (Monkeypox) Molecular Test Cartridge foil pouch when you are ready to test. Do not open the foil pouch more than 30 minutes before you begin a test. If more than 30 minutes have passed since you opened the foil pouch containing the Cartridge, obtain a new Cartridge.
- Do not use scissors or sharp objects to open the foil pouch as damage to the contents can occur.
- The Cue Sample Wand is sterile. Do not use if the packaging is damaged or accidentally opened before use. Open another Cartridge foil pouch for a sterile Cue Sample Wand.
- If the Cue Mpox (Monkeypox) Molecular Test Cartridge or Sample Wand is dropped, cracked, or found to be damaged when opened, do not use. Discard the damaged Cartridge and/or Wand and open another Cartridge foil pouch for a new Cartridge and/or sterile Sample Wand.
- Store and use the Cue Mpox (Monkeypox) Molecular Test Cartridge at the temperatures provided in the storage and testing conditions sections in these instructions for use (15-30°C).
- The Cue Mpox (Monkeypox) Molecular Test Cartridge will heat up inside the Cue Reader for one minute. Insert the Cue Sample Wand with the sample when the Cue Health App screen shows that the Cartridge heat cycle is complete. Do not wait longer than 10 minutes after the heat cycle is complete to insert the Wand.
- After the test is complete, remove the Cue Mpox (Monkeypox) Molecular Test Cartridge with the

Cue Sample Wand still inside and dispose per institutional waste guidelines. Treat all biological specimens, including used Cartridges with Wands, as if capable of transmitting infectious agents.
- Do not attempt to open the Cue Mpox (Monkeypox) Molecular Test Cartridge.
- Do not remove the Wand from the Cue Mpox (Monkeypox) Molecular Test Cartridge after it has been inserted.

Precautions – Lesion Swab Collection

- Specimen collection must occur by trained healthcare personnel.
- Do not clean the lesion with ethanol or any other disinfectant prior to swabbing.
- For best results, swab a lesion that is in one of the following stages: papular (solid and raised with distinct borders), vesicular (a lesion with fluid trapped under the skin), or pustular (a lesion that is filled with yellow fluid). Lesions that look flat, like a rash (macular), may also be swabbed.
- You must insert the Sample Wand with the skin lesion sample into the Cue Mpox (Monkeypox) Molecular Test Cartridge within 5 minutes of collecting the lesion sample.

Limitations

- Detection of monkeypox virus DNA is dependent on the number of copies present in the specimen. Detection of monkeypox nucleic acid may be affected by sample collection methods (e.g., if a specimen is improperly collected, transported, or handled), sample storage, patient factors (e.g., presence, type, and duration of symptoms), and/or stage of infection (e.g., if collected too early or too late in the course of illness), and/or interfering substances.
- The Cue Mpox (Monkeypox) Molecular Test has been evaluated only for use in combination with the following external controls – External Positive Control NATMPXV-ERC and External Negative Control NATSARS(COV2)-NEG (ZeptoMetrix (Buffalo, NY)) using the procedure provided in these instructions for use only. Modifications to this procedure may alter the performance of the test.
- There is a chance that the test may give a negative result even if monkeypox viral DNA is present (false negative). A false negative result can occur when a sample is not collected or handled properly, degradation of the viral DNA during storage, not enough sample was collected, and/or mutations in the monkeypox virus.
- As with any molecular test, mutations within the target regions of the Cue Mpox (Monkeypox) Molecular Test could affect primer and/or probe binding, resulting in failure to detect the presence of viral DNA.
- This test cannot rule out diseases caused by other bacterial or viral pathogens.
- Negative results do not preclude monkeypox virus clade I/II infection and should not be used as the sole basis for treatment or other patient management decisions. Collection of multiple specimens (and specimens collected at different time points) from the same patient may be necessary to detect the virus.
- Viral nucleic acids may persist in vivo, independent of virus viability. Detection of viral nucleic acids does not imply that the corresponding viruses are infectious or are the causative agents for clinical symptoms.
- There is a chance that the test can give a positive result when monkeypox viral DNA is not actually present (false positive). A false positive result can occur from contamination during specimen handling or preparation, or between patient specimens if the Cartridge Reader is not properly cleaned.
- The performance of this test was established using direct lesion swab specimens collected using the Cue Sample Wand and by using Wands dipped in viral transport media (VTM) containing

previously collected lesion swab specimens only. Assay performance has not been evaluated for use with other collection media and/or specimen types. Use of other collection media and/or specimen types may lead to a false positive, false negative, or invalid result.

- The performance of this test was established based on the evaluation of a limited number of clinical specimens. The clinical performance has not been established with all circulating variants but is anticipated to be reflective of the prevalent variants in circulation at the time and location of the clinical evaluation. Performance at the time of testing may vary depending on the variants circulating, including newly emerging strains of monkeypox virus and their prevalence, which may change over time. Results from the Cue Mpox (Monkeypox) Molecular Test should be interpreted in the context of other available laboratory and clinical data.
- Results from the Cue Mpox (Monkeypox) Molecular Test should be interpreted by a trained professional in conjunction with the patient's history and clinical signs/symptoms and epidemiological risk factors.
- The effect of interfering substances has only been evaluated for those listed within the labeling. Interference by substances other than those described could lead to erroneous results.
- This test has been evaluated for use with human specimen material only.
- The test performance was established during the 2022 mpox outbreak in the US. The performance may vary depending on the prevalence and population tested.
- This test is a qualitative test and does not provide the quantitative value of detected organism present.
- This test has not been evaluated for patients without signs and symptoms of monkeypox virus clade I/II.
- This test has not been evaluated for monitoring treatment of monkeypox virus clade I/II infection.
- The impacts of specific vaccines, antiviral therapeutics, antibiotics, chemotherapeutic or immunosuppressant drugs on the performance of this test have not been evaluated.
- This test has not been evaluated for screening of blood or blood products for the presence of monkeypox virus clade I/II.

Conditions of Authorization

The Cue Mpox (Monkeypox) Molecular Test Letter of Authorization along with authorized Fact Sheet for Healthcare Providers, the authorized Fact Sheet for Patients and authorized labeling are available on the FDA website:
https://www.fda.gov/medical-devices/emergency-use-authorizations-medical-devices/monkeypox-emergency-use-authorizations-medical-devices

To assist clinical laboratories and/or Patient Care Settings using the Cue Mpox (Monkeypox) Molecular Test (referred to in the Letter of Authorization as "Your Product"), the relevant Conditions of Authorization are listed below.

- Authorized laboratories[a] that receive the Cue Mpox (Monkeypox) Molecular Test must notify the relevant public health authorities of their intent to run your product prior to initiating testing.
- Authorized laboratories using the Cue Mpox (Monkeypox) Molecular Test must have a process in place for reporting test results to healthcare providers and relevant public health authorities, as appropriate.
- Authorized laboratories using the Cue Mpox (Monkeypox) Molecular Test must include with test result reports all authorized Fact Sheets. Under exigent circumstances, other appropriate methods for disseminating these Fact Sheets may be used, which may include mass media.
- Authorized laboratories using the Cue Mpox (Monkeypox) Molecular Test must use the Cue Mpox (Monkeypox) Molecular Test as outlined in the authorized labeling. Deviations from the authorized procedures, including the authorized instruments, authorized extraction methods, authorized clinical specimen types, authorized control materials, authorized other ancillary reagents and authorized materials required to use the Cue Mpox (Monkeypox) Molecular Test are not permitted.

- Authorized laboratories must have a process in place to track adverse events and report to Cue Customer Technical Support 833-283-8378 and to FDA pursuant to 21 CFR Part 803.
- All operators using the Cue Mpox (Monkeypox) Molecular Test must be appropriately trained in performing and interpreting the results of the Cue Mpox (Monkeypox) Molecular Test, use appropriate personal protective equipment when handling the Cue Mpox (Monkeypox) Molecular Test, and use the Cue Mpox (Monkeypox) Molecular Test in accordance with the authorized labeling.
- Cue Health Inc., authorized distributors, and authorized laboratories must collect information on the performance of the Cue Mpox (Monkeypox) Molecular Test and report any significant deviations from the established performance characteristics of the Cue Mpox (Monkeypox) Molecular Test of which they become aware to DMD/OHT7-OIR/OPEQ/CDRH (via email: CDRH-EUA Reporting@fda.hhs.gov). In addition, authorized distributor(s) and authorized laboratories must report to Cue Health Inc. (833.283.8378 or support@cuehealth.com).
- Cue Health Inc., authorized distributors, and authorized laboratories using the Cue Mpox (Monkeypox) Molecular Test must ensure that any records associated with this EUA are maintained until otherwise notified by FDA. Such records will be made available to FDA for inspection upon request.

[a] The letter of authorization refers to "authorized laboratories" as follows: Testing on the Cue Health Monitoring System (Cue Reader) and Cue Cartridge is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA) 42 U.S.C. §263a, that meet requirements to perform high, moderate, or waived complexity tests. Testing is authorized for use at the Point of Care (POC), i.e., in patient care settings operating under a CLIA Certificate of Waiver, Certificate of Compliance, or Certificate of Accreditation.

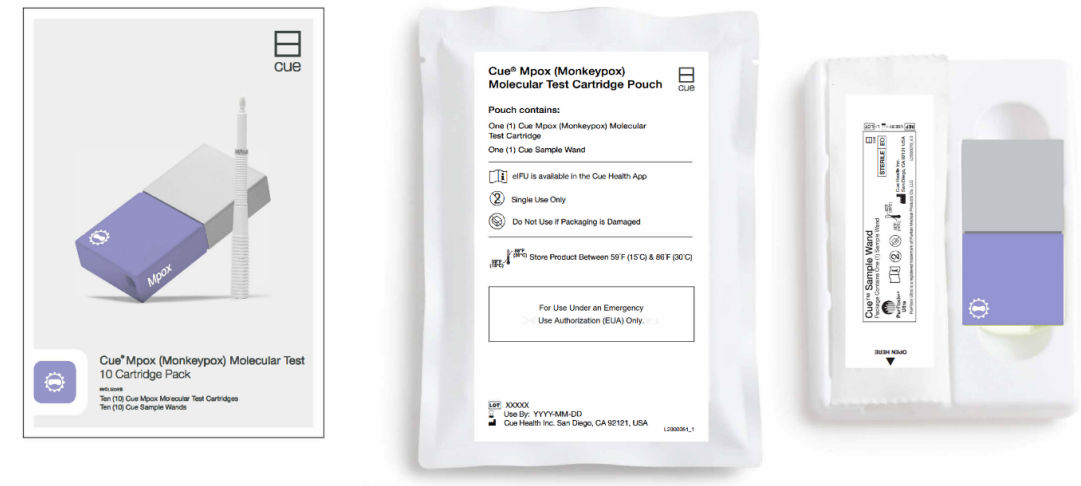
Materials Provided

- **Cue Mpox (Monkeypox) Molecular Test Cartridge Pack**

Each Cue Mpox (Monkeypox) Molecular Test Cartridge Pack contains one or more foil pouches.

- REF C3090-10 (10 Pack, contains 10 foil pouches with 10 test Cartridges and 10 Sample Wands)

Each foil pouch contains a plastic tray with one (1) single-use Cue Mpox (Monkeypox) Molecular Test Cartridge and one (1) single-use wrapped sterile Cue Sample Wand.



A small pouch called a desiccant is under the Cartridge. This pouch has material inside to protect the Cue Mpox (Monkeypox) Molecular Test Cartridge from damage due to humidity. Throw away the desiccant after the Cartridge is used.

Contact Cue Health Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378) if any component is missing or damaged, or if a Cartridge foil pouch is not sealed. You may also contact Cue Health Customer Support to request a physical copy of the Instructions For Use and the Quick Reference Instructions, free of charge.

Materials Required but Not Provided

- **Cue Health Monitoring System**
  Purchase the Cue Health Monitoring System (Cue Reader) (REF C0201) from Cue Health Inc., by contacting Cue Health Customer Support support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378).
- **Mobile Smart Device**
  Go to www.cuehealth.com for the list of compatible mobile smart devices. BLUETOOTH® wireless technology and Wi-Fi® or cellular capability is required to download the Cue Health App.
- **Cue Health Application installed on the mobile smart device**
  Download the Cue Health App from the Apple® App Store® or Google Play™ Store.
- **External Controls**
  Controls for the Cue Mpox (Monkeypox) Molecular Test are available from ZeptoMetrix. Catalog number NATMPXV-ERC (Positive Control) and Catalog number NATSARS(COV2)-NEG (Negative Control) at www.zeptometrix.com or by calling (800) 274-5487.
- Samco Exact Volume Transfer Pipette (Thermo Scientific, Product Number 966NL)
- ARX Sciences Viral Transport Media Collection Kit (Fisher Scientific, Cat # 23-444-014)

Cue Mpox (Monkeypox) Molecular Test Cartridge Storage Conditions

Store the unopened Cue Mpox (Monkeypox) Molecular Test Cartridge Pack and the foil pouches inside the pack in the temperature range shown in the table below. Do not use a Cartridge that has been stored outside of this temperature condition.

Storage Temperature	59°F (15°C) to 86°F (30°C)

Do not use a Cartridge beyond the Use By date on the Cartridge foil pouch label.

Cue Mpox (Monkeypox) Molecular Test Testing Conditions

Run a Cue Mpox (Monkeypox) Molecular Test in the temperature range shown in the table below. Do not run the Cue Mpox (Monkeypox) Molecular Test if you are outside of this temperature condition. Use caution if using this device outdoors as it has not been tested at extreme high or low temperatures or high humidity.

Operational Temperature	59°F (15°C) to 86°F (30°C)

Quality Control (QC)

External Positive and Negative Controls

External Positive and Negative Controls should show that the Cue Mpox (Monkeypox) Molecular Test is working properly. Controls for the Cue Mpox (Monkeypox) Molecular Test are available from

ZeptoMetrix. Catalog number NATMPXV-ERC (Positive Control) and Catalog number NATSARS(COV2)-NEG (Negative Control) at www.zeptometrix.com or by calling (800) 274-5487. Controls should be stored according to the manufacturer's instructions.

Cue Health recommends that negative and positive controls be run:

- With each new lot of Cartridges received
- When problems with testing are suspected or identified
- As deemed necessary in order to conform with your internal quality control procedures, with local, state and/or federal regulations, or accrediting groups

Preparation of the Controls for Use

Follow the manufacturer's instructions for mixing the controls.

Negative Control

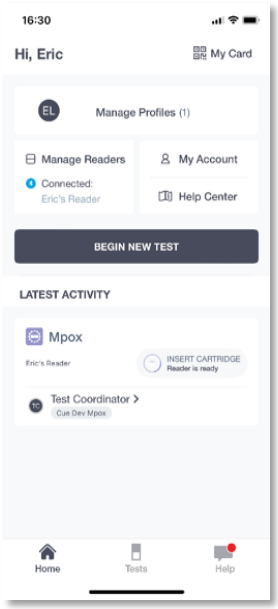
Negative Control is supplied ready to use and does not need further processing before use.

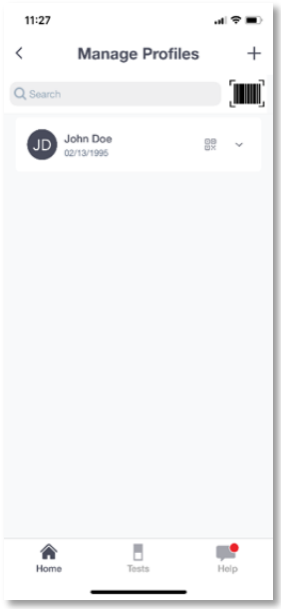
Positive Control

Positive Control needs to be diluted using a 300 µL Samco Exact Pipette (Product Number 966NL). Pipette 300 µL of the Positive Control into 3 mL of Viral Transport Media (VTM) (Fisher Scientific, Cat # 23-444-014). Invert the tube to mix.

The instructions for dipping into the Negative Control vial and the diluted Positive Control vial are: (1) insert the Sample Wand tip into the liquid and swirl the Wand as you rotate around the inner walls of the vial 3 times; (2) lift the Wand tip above the liquid level and swirl the Wand tip 3 times against the inside of the vial to dispel air bubbles; (3) place the Wand tip back into the liquid and again swirl the Wand as you rotate around the inner walls of the vial 3 more times; (4) remove the Sample Wand from the vial.

Tap on "Manage Profiles", then tap on the barcode icon on the Dashboard screen and add a sample ID for both the Negative and Positive Control. Dip a new Cue Sample Wand into the Negative Control and test that Sample Wand in a new Cue Mpox (Monkeypox) Molecular Test Cartridge. Dip a new Cue Sample Wand into the Positive Control and test that Sample Wand in a new Cue Mpox (Monkeypox) Molecular Test Cartridge.





The expected result for the Negative Control is “Negative” and the expected result for a Positive Control is “Positive”. If correct control results are not obtained, repeat the testing using a control sample on a new Cue Sample Wand and a new test Cartridge. If the control testing continues to fail, do not perform additional clinical specimen tests or report results. Contact Cue Health Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378) before testing additional clinical specimens.

Specimen Collection and Handling

Direct Skin Lesion Sample Collection

- Specimen collection must occur by trained healthcare personnel.
- Do not clean the lesion with ethanol or any other disinfectant prior to swabbing.
- For best results, swab a lesion that is in one of the following stages: papular (solid and raised with distinct borders), vesicular (a lesion with fluid trapped under the skin), or pustular (a lesion that is filled with yellow fluid). Lesions that look flat, like a rash (macular), may also be swabbed.
- Using the Cue Sample Wand, swab vigorously back and forth on the lesion surface at least 5 times for about 5 seconds.
- Rotate the Cue Sample Wand by one-quarter to one-half turn and swab the same lesion vigorously at least 3 more times.
- Apply firm pressure when swabbing. This may result in discomfort or slight pain, but it is necessary to obtain an adequate sample.
- If the lesion ruptures while swabbing, collect some lesion fluid with the Wand.

Previously Collected Skin Lesion Sample in Viral Transport Media

- The Cue Sample Wand may be dipped into a tube containing a previously collected skin lesion sample in viral transport media (VTM). Instructions for Cue Sample Wand dipping are provided in Step 5-8.
- Lesion swab specimens collected in VTM can be stored refrigerated (2-8°C) up to seven days or at -20°C for up to 30 days before the Cue Sample Wand can be dipped and inserted into the Cartridge for testing.
    - Refer to the CDC Guidelines for Collecting and Handling Specimens for Mpox Testing https://www.cdc.gov/poxvirus/Mpox/clinicians/prep-collection-specimens.html

The Cue Sample Wand containing the skin lesion sample must be inserted into the Cartridge within 5 minutes of sample collection.

Directions for Running the Cue Mpox (Monkeypox) Molecular Test

Follow the step-by-step instructions provided below.

Step 1: Obtain Items Required but Not Provided in the Cartridge Pack

You will need the items below to run the Cue Mpox (Monkeypox) Molecular Test. These items are not included in the Cue Mpox (Monkeypox) Molecular Test Cartridge Pack.

- Cue Health Monitoring System. You can purchase the system from Cue Health Inc., by contacting Cue Health Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378).
- Go to www.cuehealth.com for the list of compatible mobile smart devices. BLUETOOTH wireless technology and Wi-Fi® or cellular capability is required to download the Cue Health App.
- The Cue Health App installed on your mobile smart device. Download the Cue Health App from Apple App Store or Google Play Store.

Step 2: Set Up Your System

Read the Cue Health Monitoring System Quick Start Guide and the User Manual before you run a Cue Mpox (Monkeypox) Molecular Test. The Quick Start Guide will help you quickly set up your Cue Health Monitoring System (Cue Reader) and get ready to run a test. The User Manual gives you all the information you need to use your Cue Health Monitoring System correctly and safely. The Quick Start Guide or the User Manual will show you step-by-step how to do the following:

1. Unpack and set up the Cue Cartridge Reader.
2. Download the Cue Health App by going to the Apple App Store or Google Play Store and searching for the Cue Health App.
3. Set up your Cue Account in the Cue Health App. Once you have set up a Cue Account, you may create and edit account profiles for persons being tested. All your test data will be saved under your Cue Account in the Cue Health App and on the Cue Health secure cloud server.
4. Pair Cue Reader(s) to your mobile smart device.
5. Connect the Cue Health App to a paired Cue Cartridge Reader to run a Cue Mpox (Monkeypox) Molecular Test.
6. Learn more about your Cue Health Monitoring System and all the above system set-up steps in the Cue Health Monitoring System User Manual.

Step 3: Review All Information

Review the information provided in this Cue Mpox (Monkeypox) Molecular Test Instructions for Use before running a test. If you do not understand the instructions, do not run a test. Contact Cue Health

Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378) for help.

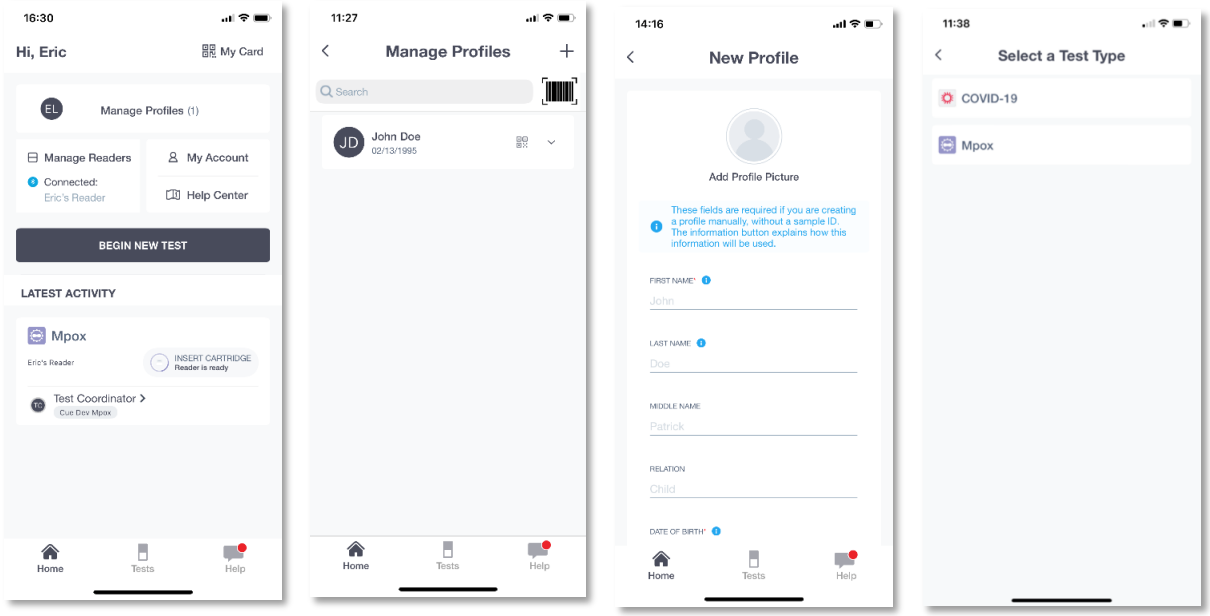
The Cue Health App uses pictures and videos to walk you through, step-by-step, how to collect a skin lesion sample and run a Cue Mpox (Monkeypox) Molecular Test. If you do not follow the instructions, the test may not run as it should, and you may not receive a test result or the test result may not be correct.

Step 4: Open the Cue Health App on Your Mobile Smart Device and Follow the On-Screen Instructions

1. The first time you use the Cue Health App you must accept the "Terms of Use and End User License Agreement" and the "Privacy Policy."
2. A Cue Health App update may be required before you run a test. Follow any on-screen instructions for updating the Cue Health App.
3. The first time you use the Cue Health App you will need to tap Create Account. After Creating an Account you may Login.
4. Make sure that the Cue Cartridge Reader you will be using is paired to your mobile smart device. Follow the Cue Health Monitoring System's Quick Start Guide or User Manual and the on-screen instructions to pair the Cue Cartridge Reader to the mobile smart device.
5. Make sure that the Cue Health App is connected to the Cue Cartridge Reader that you will be using for the Cue Mpox (Monkeypox) Molecular Test. Follow the Cue Health Monitoring System User Manual and the on-screen instructions to connect to the Cue Cartridge Reader.
6. Follow the on-screen instructions to run a test. Step 5 below also tells you how to run a test using the Cue Health App.

Step 5: Run a Cue Mpox (Monkeypox) Molecular Test

Log into your Cue Account. After logging into your account, tap on Manage Profiles. Choose the person's name or barcode ID being tested or add a new profile. To add a new profile, tap the + sign, type in the person's identification information and SAVE. Tap the barcode icon to scan a patient barcode ID. Tap on the name or patient barcode ID. Then tap "BEGIN NEW TEST" and you will see the screen below to choose to run the Cue Mpox (Monkeypox) Molecular Test.



If prompted, select your organization or select “NOT TESTING FOR AN ORGANIZATION.”

The instructions below are the same step-by-step instructions as shown in the Cue Health App videos and screens.

REMINDER: If your mobile smart device loses battery charge while performing the test, the test on the Cue Cartridge Reader will still run to completion. The test result will be saved. The mobile smart device must be charged to see the test result. Make sure your mobile smart device is close to the Cue Reader after a test completes so you can view the result on the screen in the Cue Health App.

Step 5-1: View Intended Use
Read the Intended Use presented to you in the Cue Health App and then you may continue.

Step 5-2: View Precautions
Precautions are important to follow to ensure that the test runs correctly. Limitations are important to understand before you run the test. See the Precautions and Limitations sections in this document.

Wear personal protective equipment when collecting patient samples and when running the Cue Mpox (Monkeypox) Molecular Test.

Read the precautions presented to you in the Cue Health App and then you may continue.

Step 5-3: Pair the Cue Health App to the Cue Cartridge Reader(s)
Connect the Cue Cartridge Reader to power by using the Cue Power Adapter and Cue Charging Cable. Follow the Cue Health App videos and screen instructions to pair the Cartridge Reader that will be used for the test(s) to your mobile smart device. When a paired Cartridge Reader is within BLUETOOTH wireless technology range of the mobile smart device, the Cartridge Reader is “connected” to the Cue Health App. The same instructions are in the Quick Start Guide and the Cue Health Monitoring System User Manual.

Step 5-4: Gather the Materials to Run the Test
Place the Cue Reader and a Cue Mpox (Monkeypox) Molecular Test Cartridge foil pouch in front of you. See the Cue Health App video showing these materials that you need to run a test as shown in Figure 5-4.

> **REMINDER:** Open the Cue Mpox (Monkeypox) Molecular Test Cartridge foil pouch when you are ready to test. Do not open the foil pouch more than 30 minutes before you begin a test. If more than 30 minutes have passed since you opened the foil pouch containing the Cartridge, obtain a new Cartridge.

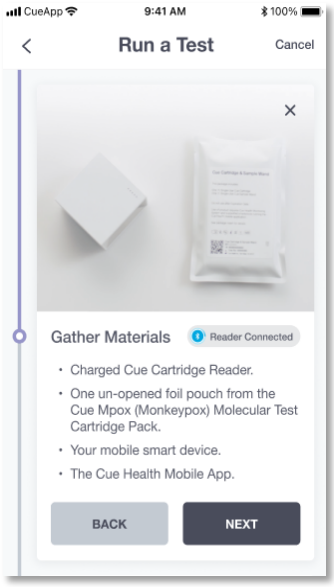


Figure 5-4

Step 5-5: Prepare the Cue Mpox (Monkeypox) Molecular Test Cartridge and Cue Sample Wand for a Test

Tear open the top of the Cartridge foil pouch and remove the plastic tray with the Cue Mpox (Monkeypox) Molecular Test Cartridge and sterile Cue Sample Wand. Remove the Cue Mpox (Monkeypox) Molecular Test Cartridge and the wrapped Cue Sample Wand from the tray.

See the Cue Health App video showing how to prepare the Cue Mpox (Monkeypox) Molecular Test Cartridge and Cue Sample Wand for a test as shown in Figure 5-5.

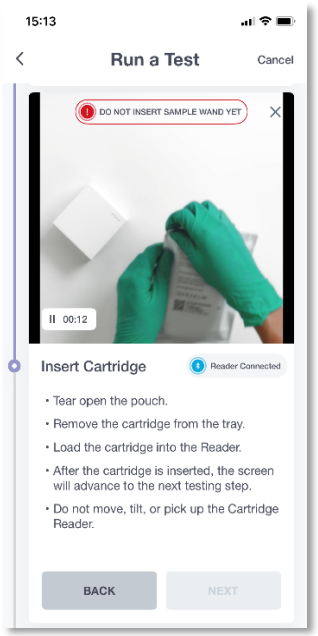


Figure 5-5

Step 5-6: Insert the Cue Mpox (Monkeypox) Molecular Test Cartridge into the Cue Reader

See the Cue Health App video showing how to insert the test Cartridge into the Cue Reader as shown in Figure 5-6-1.

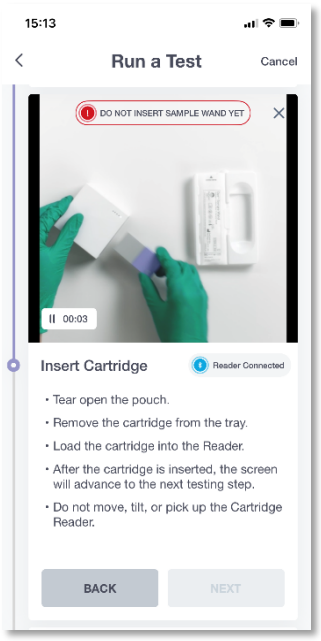


Figure 5-6-1

REMINDER: The test Cartridge must be inserted first before the Cue Sample Wand. The test Cartridge must be inserted logo side up.

REMINDER: The Cue Reader needs to be on a level surface when the Cue Mpox (Monkeypox) Molecular Test Cartridge is inserted and while the test is running. Do not move the Cue Reader while the test is running.

Support the back of the Cue Reader with one hand and hold the Cue Mpox (Monkeypox) Molecular Test Cartridge in the other hand. Insert the Cartridge (logo side up) into the Cartridge port of the Reader. When you have fully inserted the Cartridge, all five lights on top of the Cue Reader will flash and the Cue Health App screen will progress to the screen entitled “Cartridge Preheating”.

REMINDER: The test Cartridge must heat up for the full 100% heat cycle before the Cue Sample Wand is inserted into the Cartridge. All of the LED lights on the Reader will flash 5 times when the test Cartridge is ready for the Cue Sample Wand.

When you have inserted the test Cartridge all the way in, the Cartridge will start to heat up to prepare for a test, and you will see the Cue Health App video as shown in Figure 5-6-2. When the test Cartridge has finished heating up, the progress circle will show 100%.

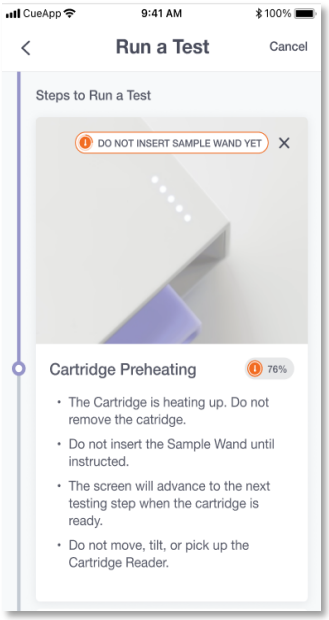


Figure 5-6-2

Step 5-7: Collect a Direct Skin Lesion Sample with the Cue Sample Wand and Insert Into the Test Cartridge

When the test Cartridge heating cycle is completed, the Cue Health App will advance to the Collect Sample screen. You may collect a direct skin lesion sample or you may dip the Cue Sample Wand into a tube containing a previously collected skin lesion sample in viral transport media (VTM).

Open the wrapped Cue Sample Wand on the side labeled "Open Here." Grasp the handle of the Cue Sample Wand and remove it from the wrapping. The Wand is sterile. Make sure the Wand tip does not touch anything. If the Wand accidentally touches any surface or is dropped, use a new Cue Sample Wand.

Proceed to Step 5-8 for instructions on dipping the Cue Sample Wand into a tube containing a previously collected skin lesion sample in viral transport media. Continue below for collecting a direct skin lesion sample.

You will see a video on how to collect a direct skin lesion sample and insert the Cue Sample Wand into the test Cartridge as shown in Figures 5-7-1 and 5-7-2.

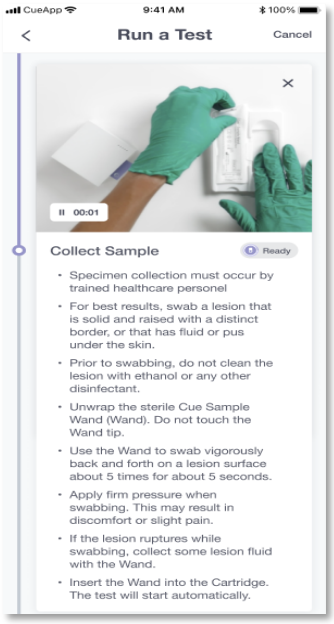


Figure 5-7-1

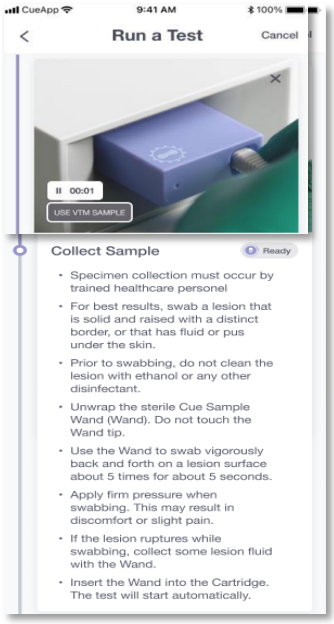


Figure 5-7-2

REMINDER: It is important to collect the skin lesion sample at the time of the Collect Sample screen and insert the Cue Sample Wand with the sample into the Cue Mpox (Monkeypox) Molecular Test Cartridge shortly after collecting the skin lesion sample. The Cue Mpox (Monkeypox) Molecular Test Cartridge should not be in the Reader without the inserted Wand for more than 10 minutes after preheating is complete, otherwise the test will automatically cancel. The Cue Health App will display reminders to insert the Cue Sample Wand as shown in Figure 5-7-3.

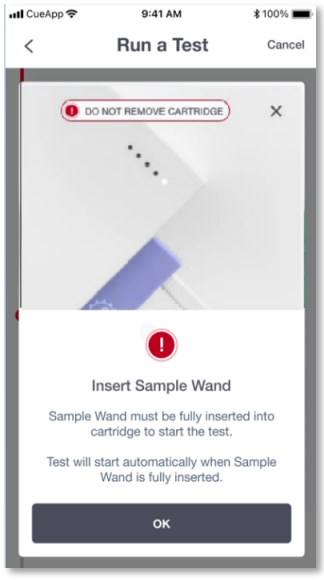


Figure 5-7-3

Follow the Cue Health App instructions below to collect a direct skin lesion swab sample with the Cue Sample Wand.

- Do not clean the lesion with ethanol or any other disinfectant prior to swabbing.
- For best results, swab a lesion that is in one of the following stages: papular (solid and raised with distinct borders), vesicular (a lesion with fluid trapped under the skin), or pustular (a lesion that is filled with yellow fluid). Lesions that look flat, like a rash (macular), may also be swabbed.
- Using the Cue Sample Wand, swab vigorously back and forth on the lesion surface at least 5 times for about 5 seconds.
- Rotate the Cue Sample Wand by one-quarter to one-half turn and swab the same lesion vigorously at least 3 more times.
- Apply firm pressure when swabbing. This may result in discomfort or slight pain, but it is necessary to obtain adequate sample.
- If the lesion ruptures while swabbing, collect some lesion fluid with the Wand.

REMINDER: You must insert the Cue Sample Wand with skin lesion sample into the Cue Mpox (Monkeypox) Molecular Test Cartridge within 5 minutes of collecting the skin lesion sample.

Support the back of the Cue Reader with one hand and hold and insert the Cue Sample Wand with skin lesion sample into the port of the Cue Mpox (Monkeypox) Molecular Test Cartridge. Make sure the Wand is inserted all the way in until the Cue Health App progresses to the screen entitled “Test in Progress”. See Step 5-9 and Figure 5-9.

Step 5-8: Sample Dipping
Skip this step if you have already collected a direct skin lesion sample with the Cue Sample Wand.

Open the wrapped Cue Sample Wand on the side labeled "Open Here." Grasp the handle of the Cue Sample Wand and remove it from the wrapping. The Wand is sterile. Make sure the Wand tip does not touch anything. If the Wand accidentally touches any surface or is dropped, use a new Cue Sample Wand.

Follow the instructions below as shown in the Cue Health App (See Figure 5-8) for dipping the Cue Sample Wand into a tube containing an individual skin lesion swab sample in viral transport media (VTM) and running the sample.

1. To ensure proper mixing, gently invert the capped VTM specimen tube, then hold the cap and gently tap the bottom of the tube on a tabletop 3 times to remove excess sample liquid from the cap.
2. Uncap the tube.
3. Insert the Cue Sample Wand tip into the liquid and swirl the Wand as you rotate around the inner walls of the tube 3 times.
4. Lift the Wand tip above the liquid level and swirl the Wand tip 3 times against the inside of the tube to dispel air bubbles.
5. Place the Wand tip back into the sample liquid and again swirl the Wand as you rotate around the inner walls of the tube 3 more times.
6. Lift the Wand tip above the liquid level and again swirl the Wand tip three times against the inside of the tube to dispel air bubbles.

7. Remove the Cue Sample Wand from the tube.
8. Support the back of the Cue Cartridge Reader and insert the Cue Sample Wand with sample into the port of the Cue Mpox (Monkeypox) Molecular Test Cartridge. Make sure the Wand is inserted all the way in until Test in Progress is shown on the Cue Health App screen. See Step 5-9 and Figure 5-9.

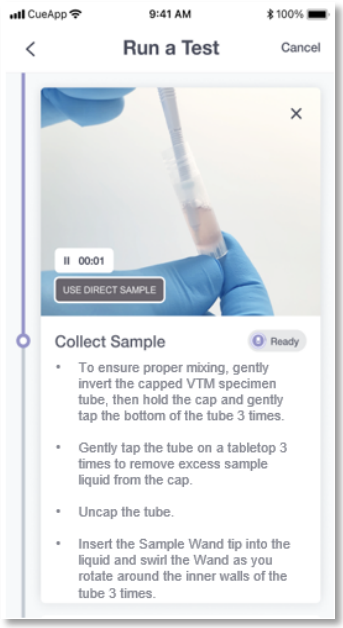


Figure 5-8

REMINDER: It is important to collect the Cue Sample Wand lesion sample by dipping into the individual sample tube at the time of the Collect Sample screen in the Cue Health App and inserting the Sample Wand with the lesion sample into the Cue Mpox (Monkeypox) Molecular Test Cartridge immediately. The Cue Mpox (Monkeypox) Molecular Test Cartridge should not be in the Cartridge Reader without the inserted Sample Wand for more than 10 minutes.

Step 5-9: Test Progress

The test will start as soon as the Cue Sample Wand is fully inserted into the Cue Mpox (Monkeypox) Molecular Test Cartridge. It takes about 25 minutes for the Cue Mpox (Monkeypox) Molecular Test to run. Once the test starts, the Cue Health App will show the test progress as percent completed as shown in Figure 5-9.

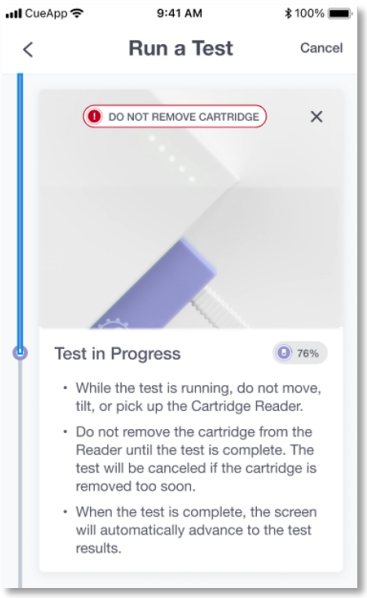


Figure 5-9

Step 5-10: View the Result

The Cue Health App will show the Cue Mpox (Monkeypox) Molecular Test result when the test is complete. The result is saved in the Cue Account profile that was selected before the test started. See Step 6 below for understanding the test results and what each result means.

Step 5-11: Remove the Cue Mpox (Monkeypox) Molecular Test Cartridge with the Cue Sample Wand After Testing

Remove the test Cartridge from the Cue Cartridge Reader by holding the Cartridge Reader with one hand and carefully pulling the Cartridge out of the Reader with the other hand. The Sample Wand must not be removed and should remain inside the Cartridge. Used Cartridges should be considered capable of transmitting infectious agents requiring standard precautions. Follow your institution's environmental waste procedures for proper disposal of used Cartridges. If country or regional regulations do not provide clear direction on proper disposal, used Cartridges should be disposed per WHO [World Health Organization] medical waste handling and disposal guidelines.

Step 6: Understand the Test Results

The Cue Health App shows the result as Negative, Positive, Invalid, or Canceled.

Step 6-1: Understanding a Negative Result

A Negative result means that the Cue Mpox (Monkeypox) Molecular Test did not detect monkeypox virus DNA in the sample. It is unlikely that the person you tested has mpox.

- There is a small chance that the test may give a negative result even if the person who was tested has mpox (false negative). A negative result does not rule out mpox and should not be used as the sole basis for treatment or patient management decisions.
- When diagnostic testing is negative, the possibility of a false negative result should be considered in the context of a patient's recent exposures and the presence of

clinical signs and symptoms consistent with mpox. The possibility of a false negative result should especially be considered if the patient's recent exposures or clinical presentation indicate that mpox is likely, and diagnostic tests for other causes of illness (e.g., other respiratory illness) are negative. If mpox is still suspected based on exposure history together with other clinical findings, re-testing should be considered in consultation with public health authorities.

- Risks to a patient of a false negative include delayed or lack of supportive treatment; lack of monitoring of infected individuals and their household or other close contacts for symptoms, resulting in increased risk of spread of mpox within the community; and other unintended adverse events.

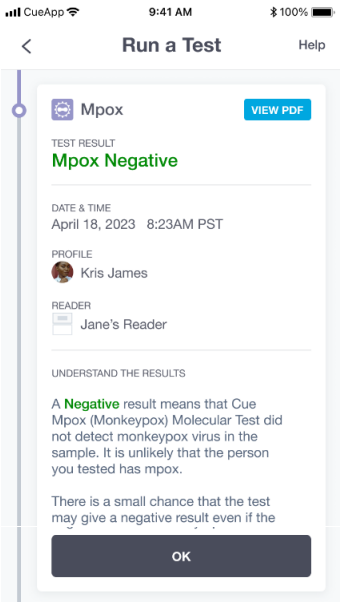


Figure 6-1

Click on back < at the top left of the Cue Health App Screen to return to a screen where you can run a new test.

Step 6-2: Understanding a Positive Result

A Positive result means that the Cue Mpox (Monkeypox) Molecular Test detected monkeypox virus DNA in the sample. It is likely that the person you tested has mpox.

- There is a small chance that this test can give a positive result even when the person tested does not have mpox (false positive). The possibility of a false positive result should be considered in the context of a patient's recent exposures and the presence of clinical signs and symptoms consistent with mpox.
- A positive result does not mean that the patient's symptoms are from mpox alone. It is possible that another type of illness is contributing to their symptoms.
- Risks to a patient of a false positive includes an unnecessary recommendation for the patient to isolate; unnecessary monitoring of household or other close contacts for symptoms; patient isolation that might limit contact with family or friends and may increase contact with mpox infected patients; limits in the ability to work; delayed diagnosis and treatment of the infection causing the symptoms; unnecessary treatment or therapy; or other unintended adverse effects.

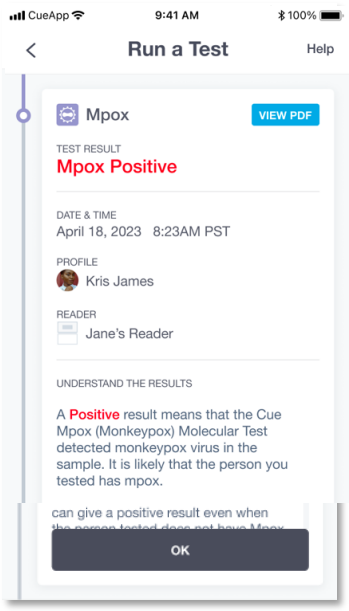


Figure 6-2

Click on back < at the top left of the Cue Health App Screen to return to a screen where you can run a new test.

Step 6-3: Understanding an Invalid Result

An Invalid result means that you may not have collected enough sample, or a system error occurred, and the Cue Health Monitoring System is not able to provide the result of the test. Retesting is required.

Common causes of invalid results are:

- You did not collect enough sample
- A processing error occurred inside the test Cartridge

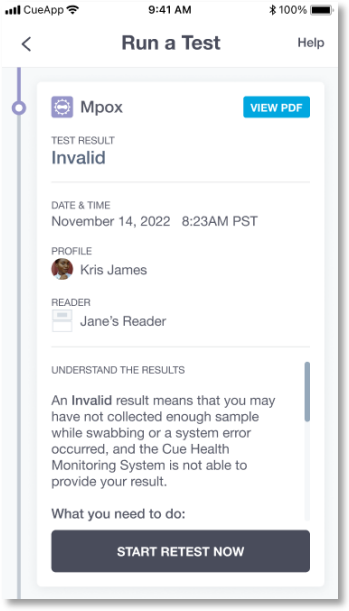


Figure 6-3

If the result is Invalid, retest. Click on START RETEST NOW. You must use a new Cue Mpox (Monkeypox) Molecular Test Cartridge and a new Cue Sample Wand.

Step 6-4: Understanding Test Result Canceled

You will see a test result of Canceled if you purposely canceled the test by tapping "Cancel" in the top right corner of the Cue Health App screen, if the system canceled the test due to a mechanical error, or because you did not follow the test instructions correctly.

Examples of when the system will cancel a test include:

- The Cue Cartridge Reader is moved or tilted while the test is running
- The test Cartridge was stored outside of recommended conditions and not equilibrated to the claimed storage range (15-30°C) prior to testing
- The test Cartridge is removed before the test is completed
- The Cue Sample Wand is inserted into the test Cartridge too soon (the Cartridge did not complete preheating) or too late (more than 10 minutes after the test Cartridge completes preheating).

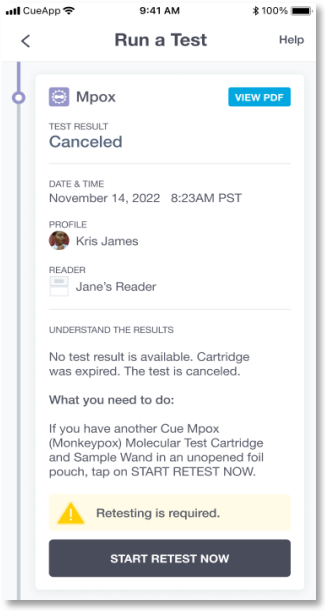


Figure 6-4

If the result is canceled, retest. Click on START RETEST NOW. You must use a new Cue Mpox (Monkeypox) Molecular Test Cartridge and a new Cue Sample Wand.

Cleaning and Disinfecting the Cue Cartridge Reader

The Cue Cartridge Reader should be cleaned and disinfected after each use. Wipe down with Clorox® Germicidal Wipes or equivalent (containing 0.55% sodium hypochlorite as the active ingredient). Do not spray any cleaning solution directly onto the Cue Cartridge Reader or into the Cartridge Port. Do not put any part of the Cue Cartridge Reader under water or any other liquid. Do not attempt to clean any internal parts.

Disposal of the Used Cue Mpox (Monkeypox) Molecular Test Cartridge

After each test, the Cue Mpox (Monkeypox) Molecular Test Cartridge with the Cue Sample Wand still inside must be removed from the Cue Cartridge Reader. Used test Cartridges should be considered

capable of transmitting infectious agents requiring standard precautions. Follow your institution's environmental waste procedures for proper disposal of used Cartridges. If country or regional regulations do not provide clear direction on proper disposal, used Cartridges should be disposed per WHO [World Health Organization] medical waste handling and disposal guidelines.

Analytical Performance

Limit of Detection (LoD)

The Limit of Detection (lowest concentration with ≥95% detection) for the Cue Mpox (Monkeypox) Molecular Test was determined with one strain of chemically inactivated monkeypox (ZeptoMetrix; USA/MA001/2022, Cat #NATMPXV-ERC). The concentration of the monkeypox virus stock was 15 copies/µL as determined by the manufacturer. The monkeypox virus was serially diluted in skin clinical matrix (SCM) which is made of negative skin swabs eluted in Phosphate Buffered Saline (PBS), to obtain four low level concentrations. Ten microliters of each dilution were applied to a dry Cue Sample Wand followed by insertion into the preheated test Cartridges for testing. The dilutions were tested in triplicate using two Cue Mpox (Monkeypox) Molecular Test Cartridge lots on each of three days for a total of 60 replicates per dilution. The confirmed LoD was one (1) genome copy/Sample Wand or 0.1 genome copies/µL of sample.

Limit of Detection Confirmation

Material	Claimed LoD Genome Copies/Sample Wand	Claimed LoD Genome Copies/µL of Sample	Confirmation Positives/Replicates
Inactivated Mpox virus (USA/MA001/2022)	1	0.1	40/40[1]

[1] Two initial tests were canceled and retested. The repeat testing generated positive results.

Performance around LoD (Reproducibility)

The performance of the Cue Mpox (Monkeypox) Molecular Test near the LoD was assessed using contrived samples that were tested in a blinded fashion by untrained operators. A total of 36 contrived positive samples consisted of inactivated monkeypox virus (ZeptoMetrix; USA/MA001/2022, Cat #NATMPXV-ERC) prepared at 1.5x LoD in skin clinical matrix (SCM). A total of 36 negative samples were made of SCM only. On each testing day, a trained laboratorian prepared and passed the Sample Wands to the untrained operator to run the test. The study was performed using three internal, simulated POC sites each with four Cartridge Readers. Each panel member was run four times per day per site over three days. Overall agreement with the expected results was 100% for contrived positive and negative samples.

Evaluation of Performance Around the Assay LoD

Operator	Day	Detected/Total Tested		% Result Agreement
		Negative Sample	Positive Sample	
1	1	0/4	4/4	100%
	2	0/4[1]	4/4	100%
	3	0/4	4/4	100%
2	1	0/4[1]	4/4	100%
	2	0/4	4/4[1]	100%
	3	0/4	4/4	100%
3	1	0/4[2]	4/4	100%
	2	0/4	4/4	100%
	3	0/4	4/4	100%

[1] One initial canceled test was retested. The repeat testing generated the expected result.
[2] One initial invalid test was retested. The repeat testing generated the expected negative result.

VTM Matrix Equivalency Study

The LoD of the Cue Mpox (Monkeypox) Molecular Test was confirmed in VTM with natural lesion swab matrix. This study also served to validate the use of skin clinical matrix (SCM), a surrogate for natural lesion swab matrix used in the original LoD study. Contrived specimens were prepared with quantified inactivated monkeypox virus (USA/MA001/2022) in pooled negative skin lesion swab samples collected in VTM at three concentrations relative to the LoD established for the dry Cue Sample Wand (i.e., 1 genome copy/Wand). Wands were prepared by applying 10 µL of inactivated monkeypox virus in pooled VTM lesion swab matrix at different concentrations onto the swab head. The confirmed assay LoD for lesion swab matrix in VTM was 1.5 genome copies/Wand which is similar to the LoD of 1 genome copy/Wand for a dry lesion Cue Sample Wand. This study also demonstrated equivalence between SCM and natural lesion swab matrix containing VTM.

Analytical Reactivity/Inclusivity

The Cue Mpox (Monkeypox) Molecular Test primers/probe were evaluated *in silico* against all available monkeypox virus strains of the Congo Basin clade (clade I) and the West African clade (clade II) in the NCBI Virus database as of February 2023. Thirty-four monkeypox virus strains of the Congo Basin clade (clade I) and 2056 West African clade (clade II) sequences were used in the analysis. The reverse primer has a single mismatch in the 5th position from the 5′ end for all clade I monkeypox virus strains that were evaluated. One of the 2056 clade II sequences exhibited a single mismatch in the 9th position from the 5′ end of the forward primer. The presence of a single mismatch is not predicted to impact test performance. The Cue Mpox (Monkeypox) Molecular Test primers/probe are predicted to be inclusive of all available monkeypox virus strains of the Congo Basin clade (clade I) and the West African clade (clade II) as of February 2023 as shown in the table below.

Percent Match of Primers/Probe to Available Clade I and Clade II Sequences as of February 2023

Primer	# of Sequences Evaluated	# of Sequences with 100% Homology	% of All Analyzed Genomes with Perfect Match
Forward Primer	2150	2149/2150	99.96%
Reverse Primer	2150	2116/2150	98.42%
Probe	2150	2150/2150	100.00%

Analytical Specificity – Cross-Reactivity *In Silico* Analysis

An *in silico* analysis for possible cross-reactivity with 10 viruses, 2 fungi, and 20 bacteria was conducted by mapping the Cue Mpox (Monkeypox) Molecular Test primers/probe sequences to the genomic sequences of each organism shown below. Of the 97 Cowpox virus BLAST results that were analyzed, 11% exhibited >80% homology to only the monkeypox specific probe, but these sequences are not expected to cross-react since there was no homology to the monkeypox virus primers. Overall, the *in silico* analysis demonstrated that there were no microorganisms for which the reference strain and BLASTn accession IDs showed ≥80% homology with all three oligonucleotide components (forward primer, reverse primer, probe) for the monkeypox virus target.

Cross-Reactivity *In Silico* Analysis

Organism	Reference Sequence Accession ID	Forward primer results at ≥80% homology	Reverse primer results at ≥80% homology	Probe results at ≥80% homology
Acinetobacter calcoaceticus	NZ_CP020000.1	No	No	No
Bacteriodes fragilis	NZ_CP069563.1	No	No	No
Camelpox virus	NC_003391.1	No	No	No
Candida albicans	NC_032089.1 NC_032090.1 NC_032091.1 NC_032092.1 NC_032094.1 NC_032095.1 NC_032096.1	No	No	No
Chlamydia trachomatis	NC_000117.1	No	No	No
Corynebacterium diphtheriae	NZ_CP025209.1	No	No	No
Corynebacterium jeikeium	NC_007164.1	No	No	No
Cowpox virus	NC_003663.2	No	No	Yes (72-92%)
Ectromelia (mousepox) virus	NC_004105.1	No	No	No
Enterococcus faecalis	NZ_ASDH00000000.1	No	No	No
Escherichia coli	NC_002695.2 NC_000913.3	No	No	No
Herpes Simplex Virus 1	NC_001806.2	No	No	No
Herpes Simplex Virus 2	NC_001798.2	No	No	No
Human Genomic DNA	NC_000001.11 NC_000002.12 NC_000003.12 NC_000004.12 NC_000005.10 NC_000006.12 NC_000007.14 NC_000008.11 NC_000009.12 NC_000010.11 NC_000011.10 NC_000012.12	No	No	No

Organism	Reference Sequence Accession ID	Forward primer results at ≥80% homology	Reverse primer results at ≥80% homology	Probe results at ≥80% homology
	NC_000013.11 NC_000014.9 NC_000015.10 NC_000016.10 NC_000017.11 NC_000018.10 NC_000019.10 NC_000020.11 NC_000021.9 NC_000022.11 NC_000023.11 NC_000024.10 NC_012920.1			
Human papilloma virus (HPV)	NC_027779.1	No	No	No
Lactobacillus acidophilus	NC_021181.2	No	No	No
Lactobacillus jensenii	NZ_CP018809.1	No	No	No
Lactobacillus vaginalis	NZ_CP104399.1	No	No	No
Molluscum contagiosum virus	NC_001731.1	No	No	No
Mycoplasma genitalium	NC_000908.2	No	No	No
Mycoplasma pneumoniae	NZ_LR214945.1	No	No	No
Neisseria gonorrhoeae	NZ_AP023069.1	No	No	No
Pseudomonas aeruginosa	NC_002516.2	No	No	No
Streptococcus pyogenes	NZ_LS483338.1	No	No	No
Streptococcus dysgalactiae subspecies equisimilis (Group C & G)	NZ_AP023394.1	No	No	No
Streptococcus agalactiae	NZ_CP012480.1	No	No	No
Streptococcus mitis	NZ_JYGP00000000.1	No	No	No

Organism	Reference Sequence Accession ID	Forward primer results at ≥80% homology	Reverse primer results at ≥80% homology	Probe results at ≥80% homology
Staphylococcus aureus	NC_007795.1	No	No	No
Staphylococcus epidermidis	NZ_CP035288.1	No	No	No
Treponema pallidum	NC_016842.1	No	No	No
Trichomonas vaginalis	None in Ref Seq Database	No	No	No
Trichophyton rubrum	None in Ref Seq Database	No	No	No
Vaccinia virus	NC_006998.1	No	No	No
Variola virus (smallpox)	NC_001611.1	No	No	No
Varicella-Zoster virus (Chickenpox)	NC_001348.1	No	No	No

Analytical Specificity – Interfering Substances

Potential assay interference from common endogenous and exogenous substances that could be associated with a lesion swab sample was assessed. Potential interferents were tested at the highest concentration likely to be found in a skin sample. Each interfering substance in negative skin clinical matrix was tested in triplicate. Each interfering substance was also tested in triplicate in the presence of inactivated monkeypox virus (ZeptoMetrix; USA/MA001/2022, Cat #NATMPXV-ERC) at 3x LoD. If any interference was observed, an additional 20 replicates were run. In the case of douche fluid, 20 fresh prepared positive Cue Sample Wands were tested with new test Cartridges from the same Cartridge lot as the initial test and 19/20 replicates generated the expected result. One of the 20 replicates had a canceled test result and upon retesting, generated the expected positive result.

None of the substances tested were found to impact performance of the Cue Mpox (Monkeypox) Molecular Test at the concentrations tested in this study.

Interfering Substances Evaluation

Substance	Substance Level	Detected/Tested		Interference at Concentration Tested (Yes/No)
		Negative Sample	Positive Sample	
Abreva	7% (w/v)	0/3	3/3*	No
Acyclovir	7 mg/mL	0/3	3/3	No
Albumin	2.2 mg/mL	0/3	3/3	No
Benadryl Cream /Ointment	7% (w/v)	0/3	3/3	No

Substance	Substance Level	Detected/Tested		Interference at Concentration Tested (Yes/No)
		Negative Sample	Positive Sample	
Biotin	3.5 µg/mL	0/3	3/3	No
Blood/EDTA	5% (v/v)	0/3	3/3	No
Carmex	7% (w/v)	0/3	3/3	No
Casein	7 mg/mL	0/3	3/3	No
Cornstarch	2.5 mg/mL	0/3	3/3*	No
Douche	7% (v/v)	0/3	22/23**	No
Feces	0.22% (w/v)	0/3	3/3	No
Female Urine	10% (v/v)	0/3	3/3*	No
Hydrocortisone Cream	7% (w/v)	0/3	3/3	No
KY Jelly	7% (w/v)	0/3	3/3	No
Lanacane	3.5% (w/v)	0/3	3/3	No
Male Urine	10% (v/v)	0/3	3/3	No
Mucin	60 µg/mL	0/3	3/3	No
Neosporin	7% (w/v)	0/3	3/3	No
Seminal Fluid	7% (v/v)	0/3	3/3	No
Vagisil Cream	1% (w/v)	0/3	3/3*	No
Zinc Oxide Ointment	7% (w/v)	0/3	3/3*	No

*One initial invalid or canceled result was retested. The repeat testing generated the expected result.
**One initial test was negative. Twenty additional samples were tested and one canceled test was retested. The repeat testing generated the expected positive result.

Clinical Performance

The performance of the Cue Mpox (Monkeypox) Molecular Test was evaluated using 60 deidentified archived clinical lesion swab samples in VTM (30 positive samples and 30 negative samples) collected from patients suspected of mpox. A trained laboratorian prepared Cue Sample Wands by dipping into the archived lesion swab specimens in VTM. Wands were then passed to the untrained user to run the test. Wands were prepared fresh on each day of testing. All of the clinical specimens were acquired from a medical institution with accompanying positive or negative results from an FDA-cleared real-time PCR assay. A total of 30 positive specimens, including four low positive specimens (4/30 = 13.3%) and 30 negative specimens were tested by the Cue Mpox (Monkeypox) Molecular Test in a simulated point of care setting by four untrained operators. Each sample was labeled with a unique study ID, blinding the untrained operator running the test to the expected sample result.

Positive percent agreement (PPA) and negative percent agreement (NPA) were determined by comparing results of the Cue Mpox (Monkeypox) Molecular Test to the known results of the FDA-cleared real-time PCR assay. The PPA and NPA for the Cue Mpox (Monkeypox) Molecular Test were both 100%. Thirty of the 30 known positive samples were positive by the candidate Cue test. One known negative was invalid in the initial Cue test as well as the Cue retest. The internal control and target sensors' values were below the assay thresholds and resulted in an invalid result. The remaining 29 known negative samples were negative by the Cue Mpox (Monkeypox) Molecular Test.

Results are presented below for the 59 evaluable samples with valid results for both the Cue Mpox (Monkeypox) Molecular Test and the comparator method.

Cue Mpox (Monkeypox) Molecular Test versus FDA-cleared Real-Time PCR Assay

Archived Samples		FDA-cleared real-time PCR assay (Comparator)	
		Positive	Negative
Cue Mpox (Monkeypox) Molecular Test	Positive	30	0
	Negative	0	29

PPA: 100% (95% CI: 88.4% - 100%)
NPA: 100% (95% CI: 88.1% - 100%)

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Customer Support

If you have questions about this test, contact Cue Health Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378).

You can purchase the Cue Health Monitoring System and Cue Mpox (Monkeypox) Molecular Test Cartridge Packs by contacting Cue Health Customer Support at support@cuehealth.com or call toll-free at 833.CUE.TEST (833.283.8378).

Symbols Used on the Product Labels

The table below describes the symbols used on the Cue Mpox (Monkeypox) Molecular Test Cartridge Pack, the Cartridge foil pouch, and the Cue Sample Wand.

SYMBOL	DESCRIPTION
IVD	In Vitro Diagnostic
	Consult Instructions for Use eIFU available on the Cue Health Mobile Application and at www.cuehealth.com
SN	Serial Number
	Do not use if seal or packaging is broken or damaged
	Storage temperature range
REF	Catalog number
	Manufacturer
	Keep dry
	Use By

cue

Cue Health Inc.
San Diego, CA 92121 USA
Email: support@cuehealth.com
Phone: 833.CUE.TEST (833.283.8378)
Website: www.cuehealth.com