MPXV AMP Kit

REF 09R06-095
53-608429/R1

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53-608429/R1

Rx ONLY
IVD

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

CUSTOMER SERVICE: 1-800-553-7042
CUSTOMER SERVICE INTERNATIONAL:
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INTRODUCTION

This Emergency Use Authorization (EUA) package insert must be read carefully prior to use and must be followed accordingly. Reliability of EUA assay results cannot be guaranteed if there are any deviations from the instructions in this package insert.

NAME

Alinity m MPXV AMP Kit

INTENDED USE

The Alinity m MPXV assay is a real-time polymerase chain reaction (PCR) test intended for the qualitative detection of DNA from monkeypox virus (clade I/II) in human lesion swab specimens (i.e., swabs of acute pustular or vesicular rash) in viral transport media (VTM) from individuals suspected of monkeypox infection by their healthcare provider. Testing is limited to laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet the requirements to perform moderate or high complexity tests.

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 (clade I/II) 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 Alinity m MPXV assay is intended for use by qualified and trained clinical laboratory personnel specifically instructed and trained in the techniques of PCR and in vitro diagnostic procedures.

The Alinity m MPXV assay is only for use under the Food and Drug Administration's Emergency Use Authorization.

SUMMARY AND EXPLANATION OF THE TEST

Monkeypox is a disease caused by the monkeypox virus (MPXV), which is a double-stranded DNA virus that belongs to the Orthopoxvirus genus in the family Poxviridae.[1] MPXV 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 MPXV, clade I (Congo Basin –Central African Region) and clade II (West Africa).[1, 2, 3] The typical presentation of monkeypox consists of fever, rash and swollen lymph nodes.[2] Generally, clade II monkeypox infections exhibit a less severe illness.[5]

MPXV was first isolated from a 9-month-old boy from the Democratic Republic of the Congo in 1970[4] and since then most monkeypox cases have been endemic in Central and West Africa.[3,5] Since early May 2022, confirmed monkeypox cases (clade IIb) have been reported in countries that have not previously reported cases of monkeypox, and continue to be reported in several endemic countries.[6-8] On July 23, 2022, the World Health Organization (WHO) determined the monkeypox outbreak to be a public health emergency of international concern (PHEIC). WHO recommends the use of a polymerase chain reaction (PCR) based test method to provide fast and accurate detection of MPXV.

BIOLOGICAL PRINCIPLES OF THE PROCEDURE

The Alinity m MPXV assay requires 2 separate assay specific kits:

- Alinity m MPXV AMP Kit; List No. 09R06-095, consisting of 2 types of multi-well assay trays. The amplification trays (AMP TRAY 1) contain unit-dose liquid PCR amplification/detection reagents and unit-dose liquid internal control (IC) in separate wells, and the activation trays (ACT TRAY 2) contain liquid unit-dose activation reagent. The intended storage condition for the Alinity m MPXV AMP Kit is -25˚C to -15˚C.
- Alinity m MPXV CTRL Kit; List No. 09R06-085, consisting of negative controls and positive controls, each supplied as liquid in single-use tubes. The intended storage condition for the Alinity m MPXV CTRL Kit is -25˚C to -15˚C.

The Alinity m MPXV assay utilizes real-time polymerase chain reaction (PCR) to amplify and detect monkeypox virus genomic DNA sequences, internal control sequences, and human genomic DNA sequences that have been extracted from clinical specimens.

The steps of the Alinity m MPXV assay consist of sample preparation, PCR assembly, amplification/detection, and result calculation and reporting. All steps of the Alinity m MPXV assay procedure are executed automatically by the Alinity m System. No intermediate processing or transfer steps are performed by the user. The Alinity m System is designed to be a random access analyzer that can perform the Alinity m MPXV assay in parallel with other Alinity m assays on the same instrument. Application parameters specific to the Alinity m MPXV assay are contained on an assay-specific application specification file, that will be distributed electronically, and loaded onto the Alinity m System.

Nucleic acid from specimens are extracted automatically on-board the Alinity m System using the Alinity m Sample Prep Kit 2, Alinity m Lysis Solution, and Alinity m Diluent Solution. The Alinity m System employs magnetic microparticle technology to facilitate nucleic acid capture, wash and elution. The resulting purified nucleic acids are then combined with the liquid unit-dose activation reagent, liquid unit-dose amplification reagents, and Alinity m Vapor Barrier Solution, and transferred by the instrument to an amplification/detection module for PCR amplification, and real-time fluorescence detection.

The Alinity m MPXV amplification reagents include primers and probes that amplify and detect an endogenous human DNA sequence (Cellular Control) as a sample validity control for sample adequacy, sample extraction and amplification efficiency. In addition, at the beginning of the Alinity m MPXV sample preparation process, a liquid unit-dose of exogenous Internal Control (plasmid DNA) from the AMP Tray is delivered into each sample preparation reaction. The Internal Control is then processed through the entire sample preparation and real-time PCR procedure along with the specimens and controls. The Cellular Control and Internal Control are both used to demonstrate assay validity.

Assay controls are tested at or above an established minimum frequency (ie, once every 24 hours) to help ensure that instrument and reagent performance remain satisfactory. During each control event, a negative control and a positive control are processed through sample preparation and PCR procedures that are identical to those used for specimens.

The possibility of nucleic acid contamination on the Alinity m System is minimized because:

- Aerosol barrier pipette tips are used for all pipetting. The pipette tips are discarded after use.
- PCR amplification and detection is carried out automatically in a sealed reaction vessel.
- Disposal of the reaction vessel is performed automatically by the Alinity m System.

For additional information on system and assay technology, refer to the Alinity m System Operations Manual, Section 3.

REAGENTS

Kit Contents

Alinity m MPXV AMP Kit (List No. 09R06-095)

The Alinity m MPXV AMP Kit is comprised of 2 types of multi-well trays: Alinity m MPXV AMP TRAY 1 and Alinity m MPXV ACT TRAY 2.

Each Alinity m MPXV AMP TRAY 1 (individually packed in a foil pouch) contains 48 unit-dose liquid amplification reagent wells and 48 unit-dose liquid IC wells. One well of each is used per test.

- Amplification reagent wells consist of synthetic oligonucleotides, DNA Polymerase, dNTPs, and 0.15% ProClin® 950 in a buffered solution with a reference dye.
- IC wells consist of linearized plasmid DNA and carrier DNA in a TE buffer containing 0.15% ProClin 950 as a preservative.

Each Alinity m MPXV ACT TRAY 2 (individually packed in a foil pouch) contains 48 unit-dose liquid activation reagent wells. One reagent well is used per test.

- Activation reagent wells consist of magnesium chloride, potassium chloride, and tetramethylammonium chloride. Preservative: 0.15% ProClin 950.

	Quantity
Σ	192 tests
Alinity m MPXV AMP TRAY 1	4 trays / 48 tests each
Alinity m MPXV ACT TRAY 2	4 trays / 48 tests each

WARNINGS AND PRECAUTIONS

IVD

For In Vitro Diagnostic Use Under the FDA Emergency Use Authorization

- For use under an Emergency Use Authorization.
- This product has not been FDA cleared or approved, but has been authorized for emergency use by FDA under an EUA for use by authorized laboratories; use by laboratories certified under CLIA, to perform moderate or high complexity tests.
- This product has been authorized only for the detection of nucleic acid from monkeypox virus, not for any other viruses or pathogens; and
- 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 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.
- Do not use beyond expiration date
- For Prescription Use Only

For safe handling of specimens refer to the U.S. CDC and WHO guidance referenced within this package insert.

Safety Precautions

The following warnings and precautions apply to:
Alinity m MPXV AMP TRAY 1.

WARNING	Hazard-determining components of labeling: 2-Methyl-4-isothiazolin-3-one
H317	May cause an allergic skin reaction.
Prevention	
P261	Avoid breathing mist / vapors / spray.
P272	Contaminated work clothing should not be allowed out of the workplace.
P280	Wear protective gloves/protective clothing/eye protection.
Response	
P302+P352	IF ON SKIN: Wash with plenty of water.
P333+P313	If skin irritation or rash occurs: Get medical advice/ attention.
P362+P364	Take off contaminated clothing and wash it before reuse.
Disposal	
P501	Dispose of contents/container in accordance with local regulations.

The following warnings and precautions apply to:
Alinity m MPXV ACT TRAY 2.

DANGER	Hazard-determining components of labeling: Tetramethylammonium chloride and 2-Methyl-4-isothiazolin-3-one
H302	Harmful if swallowed.
H316	Causes mild skin irritation[a]
H317	May cause an allergic skin reaction.
H370	Causes damage to organs.
H412	Harmful to aquatic life with long lasting effects.
Prevention	
P260	Do not breathe mist / vapors / spray.
P264	Wash hands thoroughly after handling.
P272	Contaminated work clothing should not be allowed out of the workplace.
P273	Avoid release to the environment.
P280	Wear protective gloves / protective clothing / eye protection.
Response	
P301+P312	IF SWALLOWED: Call a POISON CENTER or doctor/ physician if you feel unwell.
P302+P352	IF ON SKIN: Wash with plenty of water.
P308+P311	IF exposed or concerned: Call a POISON CENTER or doctor/physician.
P333+P313	If skin irritation or rash occurs: Get medical advice / attention.
P362+P364	Take off contaminated clothing and wash it before reuse.
Disposal	
P501	Dispose of contents / container in accordance with local regulations.

[a] Not applicable where regulation EC 1272/2008 (CLP) or OSHA Hazard Communication 29CFR 1910.1200 (HCS) 2012 have been implemented.

Important information regarding the safe handling, transport and disposal of this product is contained in the Safety Data Sheet.

Safety Data Sheets are available from your Abbott Representative.

For a detailed discussion of safety precautions during system operation, refer to the Alinity m System Operations Manual, Section 7 and Section 8.

Reagent Shipment

	Shipment Condition
Alinity m MPXV AMP Kit	On dry ice

Reagent Storage

In order to minimize damage to foil pouches, it is recommended that the Alinity m MPXV AMP TRAY 1 (AMP TRAY 1) and Alinity m MPXV ACT TRAY 2 (ACT TRAY 2) are stored in the original kit packaging. Open the foil pouch for the reagent trays just prior to loading onto the instrument. Onboard storage time begins when reagents are loaded on the Alinity m System.

	Storage Temperature	Maximum Storage Time
Unopened	-25°C to -15°C	Until expiration date
Onboard	System Temperature	12 days (not to exceed expiration date)

Reagent Handling

- Do not use reagents that have been damaged.
- Minimize contact with the surface of reagent trays during handling.

- Only load AMP TRAY 1 and ACT TRAY 2 from the same AMP Kit lot on the same Alinity m Assay Tray Carrier. Do not load AMP TRAY 1 and ACT TRAY 2 from different AMP Kit lots on the same Alinity m Assay Tray Carrier.
- The Alinity m System will track the onboard storage time of AMP TRAY 1 and ACT TRAY 2 while on the instrument. The Alinity m System will not allow the use of AMP TRAY 1 and ACT TRAY 2 if the maximum onboard storage time has been exceeded.
- For a detailed discussion of reagent handling precautions during system operation, refer to the Alinity m System Operations Manual, Section 8.

Indications of Reagent Deterioration

- Deterioration of the reagents may be indicated when a control error occurs or control values are repeatedly out of the specified ranges.
- Reagents are shipped on dry ice and stored at -25°C to -15°C upon arrival. If reagents arrive in a condition contrary to this recommendation or are damaged, immediately contact your Abbott Representative.
- For troubleshooting information, refer to the Alinity m System Operations Manual, Section 10.

INSTRUMENT PROCEDURE

The Alinity m MPXV assay application specification file must be installed on the Alinity m System prior to performing the assay.

For a detailed description of system operating instructions, refer to the Alinity m System Operations Manual, Section 5.

SPECIMEN TYPE, STORAGE, AND PREPARATION FOR ANALYSIS

Specimen Type

Human lesion swab specimens collected in VTM from individuals suspected of monkeypox infection by their healthcare provider can be used with the Alinity m MPXV assay on the Alinity m System.

Specimen Storage and Shipping

Refer to the CDC website for lesion swab VTM specimen storage and shipping information https://www.cdc.gov/poxvirus/monkeypox/clinicians/prep-collection-specimens.html.[9] Lesion swab specimens in VTM can be stored at 5°C ± 3°C for a maximum of 7 days prior to testing, or stored at -20°C or colder for a maximum of 30 days (with a maximum of 1 freeze/thaw) prior to testing.

Preparation for Analysis

Specimen can be transferred into an Alinity m Transport Tube or an Alinity m Aliquot Tube before loading onto the Alinity m System.

IMPORTANT: If present, swab should be removed from the specimens before loading onto the Alinity m System. Alternatively, transfer sample to a clean tube before loading.

If the specimen is stored frozen, it must be completely thawed prior to sample preparation.

- Thaw specimens at 15°C to 30°C or at 2°C to 8°C.

Vortex each specimen for a minimum of 2 to 3 seconds. Ensure that the contents of each tube are at the bottom after vortexing by tapping the tubes on the bench to bring liquid to the bottom of the tube.

Visually inspect each specimen.

- If layering, stratification, or precipitation is observed, continue to mix specimens thoroughly to ensure uniformity.

If needed, centrifuge specimens at 2000 *g* for 5 minutes before loading on the Alinity m System.

All specimen tubes must be labeled with specimen ID barcodes or must be identified with a specimen ID, rack ID, and position in the rack. Refer to the **Assay Procedure** section of this package insert for tube sizes and requirements for minimum sample volume and use of caps. Avoid touching the inside of the cap when opening tubes.

PROCEDURE

Materials Provided

09R06-095 Alinity m MPXV AMP Kit

Materials Required but not Provided

- 09R06-085 Alinity m MPXV CTRL Kit
- 09N12-001 Alinity m Sample Prep Kit 2
- 09N20-001 Alinity m Lysis Solution
- 09N20-003 Alinity m Diluent Solution
- 09N20-004 Alinity m Vapor Barrier Solution
- Alinity m MPXV Application Specification File
- Vortex mixer
- Calibrated pipettes capable of delivering 100 to 1000 µL
- Aerosol barrier pipette tips for 100 to 1000 µL pipettes
- Plate adapter for 384 well plates (such as Eppendorf Catalog No. 022638955)
- Centrifuge with swing plate rotor capable of accommodating the plate adapter and capable of ≥ 100 *g*

Other Optional Materials

- 09N49-010 Alinity m Transport Tube Pierceable Capped
- 09N49-011 Alinity m Transport Tubes
- 09N49-012 Alinity m Pierceable Caps
- 09N49-013 Alinity m Aliquot Tube

For information on materials required for operation of the instrument, refer to the Alinity m System Operations Manual, Section 1.

For general operating procedures, refer to the Alinity m System Operations Manual, Section 5.

For optimal performance, it is important to perform routine maintenance as described in the Alinity m System Operations Manual, Section 9.

Procedural Precautions

- Read the instructions in this package insert carefully before processing samples.
- Do not use specimens if the collection tube is damaged or if buffer has leaked from the tube. Discard unused, damaged, or leaking tubes in accordance with local, state, and federal regulations.
- When handling specimens in Alinity m Transport Tubes and Alinity m Aliquot Tubes, do not touch the top of pierceable caps to avoid cross-contamination. Specimens can contain high levels of organisms. Change gloves if they come in contact with specimen.
- Use aerosol barrier pipette tips or disposable pipettes only one time when pipetting specimens. To prevent contamination to the pipette barrel while pipetting, care should be taken to avoid touching the pipette barrel to the inside of the sample tube or container. The use of extended aerosol barrier pipette tips is recommended.
- Work area and instrument platforms must be considered potential sources of contamination.
- Ensure the Alinity m MPXV AMP TRAY 1 and the Alinity m MPXV ACT TRAY 2 are centrifuged prior to loading on the Alinity m System per instructions in the **Assay Procedure** section.
- Monitoring procedures for the presence of amplification product can be found in the Alinity m System Operations Manual, Section 9.
- To reduce the risk of nucleic acid contamination, clean and disinfect spills of specimens by including the use of a tuberculocidal disinfectant such as 1.0% sodium hypochlorite or other suitable disinfectant.
- To prevent contamination, change to new gloves before handling the Alinity m Sample Prep Kit 2, assay trays, system solutions, Integrated Reaction Unit (IRU) sleeves, and pipette tips. Also change to new gloves whenever they are contaminated by a specimen, a control, or a reagent. Always use powder-free gloves.
- The use of the Alinity m MPXV CTRL Kit is integral to the performance of the Alinity m MPXV assay. Refer to the **QUALITY CONTROL PROCEDURES** section of this package insert for details. Refer to the Alinity m MPXV CTRL Kit package insert for preparation and usage.
- The Alinity m MPXV CTRL reagents are contained in single-use tubes with pierceable caps. Avoid contamination or damage to the caps after removal from their original packaging. Discard tubes after use.

Assay Procedure

Thaw AMP TRAY 1 and ACT TRAY 2 at 15°C to 30°C or at 2°C to 8°C immediately prior to use on the Alinity m System.

Prior to loading onto the Alinity m System, Alinity m MPXV AMP TRAY 1 and ACT TRAY 2 must be centrifuged as follows:

1. Load the trays onto the plate adapter (eg, Eppendorf Catalog No. 022638955).
2. Load the plate adapter (with the trays) on a swing plate centrifuge capable of accommodating the plate adapter. Spin at 100 *g* to 800 *g* for 1 to 5 minutes to remove potential bubbles.
3. Immediately following centrifugation, carefully transfer the trays to the Alinity m Assay Tray Carriers. Take care to minimize disturbance to the trays. Load the tray carriers per Alinity m System Operations Manual, Section 5.

4. If disturbance occurs during transfer that could potentially introduce bubbles (eg, dropping, bumping, inversion of the trays), re-centrifuge the trays.
5. Proceed with the **Reagent and sample inventory management** procedure per the Alinity m System Operations Manual, Section 5.

For a detailed description of how to run an assay, refer to the Alinity m System Operations Manual, Section 5.

Prior to testing specimens, check control status. If control testing is required refer to **QUALITY CONTROL PROCEDURES** section. Controls may be tested separately or with specimens.

For preparation of samples, refer to the instructions under **SPECIMEN PREPARATION FOR ANALYSIS: Preparation for Analysis** section.

From the Create Order screen, select the assay (MPXV) being tested. The Alinity m System will track the onboard storage time of amplification reagents, controls and specimens while on the instrument. The Alinity m System will not allow the use of amplification reagents, controls or process specimens that have exceeded the allowable onboard storage time.

Specimens may be placed on the Alinity m Universal Sample Rack (sample rack) onboard the system for up to 4 hours prior to processing.

Requirements for minimum/maximum sample volumes and cap requirements for specimen tubes allowable on the Alinity m System are summarized in the table below:

Tube Type[a]	List No.	Minimum Volume Required	Maximum Volume	Cap Requirement on Instrument
Alinity m Aliquot Tube	09N49-013	0.6 mL	3.5 mL	Uncapped[b]
Alinity m Aliquot Tube with pierceable cap (09N49-012) added	09N49-013, 09N49-012	0.6 mL	3.5 mL	Capped[c]
Alinity m Transport Tube	09N49-011	1.0 mL	3.5 mL	Uncapped[b]
Alinity m Transport Tube with pierceable cap (09N49-012) added	09N49-011, 09N49-012	0.6 mL	3.5 mL	Capped[c]
Alinity m Transport Tube Pierceable Capped	09N49-010	1.0 mL	3.5 mL	Uncapped[b]
Alinity m Transport Tube Pierceable Capped	09N49-010	0.6 mL	3.5 mL	Capped[c]
Tube with 11.5 to 14.0 mm diameter		1.3 mL	2.5 mL	Uncapped[b,d]
Tube with 14.0 to 16.0 mm diameter		1.4 mL	3.5 mL	Uncapped[b,d]

[a] Refer to the Alinity m System Operations Manual, Section 4, for sample tube specifications and requirements and Section 5 for sample rack loading instructions.

[b] Avoid touching the inside of the cap when opening the tubes.

[c] Avoid touching inside of the cap when replacing or adding a new cap. Avoid touching the septum when handling the sample tubes.

[d] If beads are present in the tube they do not need to be removed prior to testing.

Place the positive and negative controls, if applicable, and the specimens into the sample rack. If used, bar codes on tube labels must face the correct orientation for scanning.

QUALITY CONTROL PROCEDURES

Negative and Positive Controls

One Alinity m MPXV Negative CTRL and one Alinity m MPXV Positive CTRL are recommended to be tested, at the minimum frequency of once every 24 hours to monitor the assay performance and Alinity m System. Valid results for all control levels must be obtained before specimen results are reported. Additional controls may be tested in accordance with local, state, and/or federal regulations or accreditation requirements and your laboratory's quality control policy.

If quality control results do not meet the acceptance criteria, refer to the Alinity m System Operations Manual, Section 10, for troubleshooting information.

A flag or message code is displayed for specimens when a control result is invalid. If the controls are invalid, all of the specimens processed following an invalid assay control must be retested.

If control results are invalid, refer to the Alinity m System Operations Manual, Section 5 for a description of quality control flags, and Section 10 for troubleshooting information.

The presence of monkeypox virus must not be detected in the negative control. Monkeypox virus detected in the negative control is indicative of contamination by other samples or by amplified product. To avoid contamination, clean the Alinity m System and repeat sample processing for controls and specimens following the Procedural Precautions in this package insert.

Monitoring procedures for the presence of amplification product can be found in the Alinity m System Operations Manual, Section 9. If negative controls are repeatedly reactive, contact your Abbott Representative.

Detection of Inhibition and/or Cell Inadequacy

A defined, consistent quantity of Internal Control (IC) is present in each PCR reaction and measured on the Alinity m System to assess amplification efficiency and to confirm that no PCR inhibitors are present in the sample. The IC is comprised of a DNA sequence unrelated to the Alinity m MPXV assay target sequences.

The Alinity m MPXV assay also detects an endogenous human DNA sequence (CC: Cellular Control) from the specimen to evaluate sample adequacy, sample extraction, and amplification efficiency.

A Flag or Message Code is displayed when the IC CN value or the CC CN value of a specimen or control exceeds the established range:

- For Positive Specimens: If the IC CN or CC CN is out of range, but the analyte in that sample is Positive, the sample will yield a Positive result. An IC Flag or CC Flag will be reported next to the positive result.
- For Negative Specimens: If the IC CN or CC CN is out of range and the analyte in that sample is not Positive, no result will be reported for the analyte and a Message Code will be generated. The specimen should be retested if this occurs.

Refer to the Alinity m System Operations Manual, Section 5 for an explanation of the corrective actions for Flags.

Refer to the Alinity m System Operations Manual, Section 10 for an explanation of the corrective actions for Message Codes.

RESULTS

Calculation

The Alinity m MPXV assay is a qualitative assay. The amplification cycle number (CN) is determined when the fluorescent signal is detected above a specified threshold by the Alinity m System. The Alinity m System will automatically report a result and interpretation for each specimen. If applicable a message code or flag will be displayed.

Interpretation of Results

Result	Interpretation
MPXV Negative	MPXV Target Not Detected
MPXV Positive, xx.xx CN[a]	MPXV Target Detected

[a] Cycle number (CN) reported for the positive MPXV result represents the total number of PCR cycles, including the 5 initial cycles where fluorescence signal is not captured and the subsequent cycles where fluorescence signal is captured.

Flags, Result Codes, and Message Codes

Some results may contain information in the Flags and Codes fields. For a description of the flags and result codes that may appear in these fields, refer to the Alinity m System Operations Manual, Section 5.

For a description of Message Codes refer to the Alinity m System Operations Manual, Section 10.

LIMITATIONS OF THE PROCEDURE

- **For use under an Emergency Use Authorization only.**
- This assay is for in vitro diagnostic use under FDA Emergency Use Authorization only.
- Use of the Alinity m MPXV assay is limited to personnel who have been trained in the procedures of a molecular diagnostic assay and the Alinity m System.
- Laboratories are required to report all results to the appropriate public health authorities.
- The instruments and assay procedures reduce the risk of contamination by amplification product. However, nucleic acid contamination from the positive controls or specimens must be controlled by good laboratory practice and careful adherence to the procedures specified in this package insert.
- Optimal performance of this test requires appropriate specimen collection and handling (refer to the **SPECIMEN TYPE, STORAGE, AND PREPARATION FOR ANALYSIS** section of this package insert).
- This assay does not detect variola virus (smallpox virus).

- While monkeypox virus clade II is the only member of the Orthopoxvirus genus known to be circulating among humans in the US at this time, a positive result most likely represents the presence of monkeypox virus clade II, although there is a small possibility that this result could represent the presence of monkeypox virus clade I. If clinical concern for such an infection exists, healthcare providers should contact the CDC and their local public health authorities for guidance.
- Performance of the assay has been evaluated using contrived clinical lesion swab specimens. Clinical Performance with natural clinical lesion specimens has not been established.
- The assay is indicated for testing of lesion swab specimens. Performance for other specimen types has not been established.
- Performance of the test has only been established in lesion swabs collected in VTM. Performance of the test has not been evaluated for dry swabs or for lesion swabs collected in other transport media types.
- A specimen with a result of "MPXV Negative" does not preclude monkeypox virus 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.
- Detection of monkeypox virus DNA may be affected by sample collection methods (eg, if a specimen is improperly collected, transported, or handled), patient factors (eg, presence, type, and duration of symptoms), and/or stage of infection (eg, if collected too early or too late in the course of illness).
- False-negative results may arise from degradation of the viral DNA during storage and transport of the specimens.
- Interfering substances studies have not been performed for this assay. The assay uses conventional well-established nucleic acid extraction methods used for other similar assays. Interference from common endogenous substances is not anticipated.
- The impacts of specific vaccines, antiviral therapeutics, antibiotics, chemotherapeutic or immunosuppressant drugs on the performance of this test have not been evaluated.
- As with any molecular test, mutations within the target regions of Alinity m MPXV assay could affect primer and/or probe binding resulting in failure to detect the presence of virus.
- 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 change over time.
- Results should be interpreted by a trained professional in conjunction with the patient's history and clinical signs and symptoms, and epidemiological risk factors.

CONDITIONS OF AUTHORIZATION FOR LABORATORIES

The Alinity m MPXV assay Letter of Authorization, along with the 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.

However, to assist clinical laboratories using the Alinity m MPXV assay ("your product" in the conditions below), the relevant Conditions of Authorization are listed below:

A. Authorized laboratories[a] using your product 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.

B. Authorized laboratories[a] using your product must use your product 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 your product are not permitted.

C. Authorized laboratories[a] that receive your product must notify the relevant public health authorities of their intent to run your product prior to initiating testing.

D. Authorized laboratories[a] using your product must have a process in place for reporting test results to healthcare providers and relevant public health authorities, as appropriate.

E. Authorized laboratories[a] must have a process in place to track adverse events and report to you (via email: molecularsupport@abbott.com; 1-800-553-7042) and to FDA pursuant to 21 CFR Part 803.

F. All laboratory personnel using your product must be appropriately trained in real time PCR techniques and use appropriate laboratory and personal protective equipment when handling this kit, and use your product in accordance with the authorized labeling.

G. Abbott, authorized distributor(s) and authorized laboratories[a] must collect information on the performance of your product and must report any significant deviations from the established performance characteristics of your product of which they become aware to DMD/OHT7/OPEQ/CDRH (via email: CDRH-EUAReporting@fda.hhs.gov). In addition, authorized distributor(s) and authorized laboratories report to Abbott (via email: molecularsupport@abbott.com; 1-800-553-7042).

H. Abbott, authorized distributor(s) and authorized laboratories[a] must ensure that any records associated with this EUA are maintained until otherwise notified by FDA. Such records must be made available to FDA for inspection upon request.

[a] The letter of authorization refers to "Laboratories certified under the Clinical Laboratory Improvement Amendments of 1988 (CLIA), 42 U.S.C. §263a, that meet the requirements to perform moderate or high complexity tests" as "authorized laboratories."

SPECIFIC PERFORMANCE CHARACTERISTICS

Limit of Detection (Analytical Sensitivity)

Limit of Detection (LoD) studies determine the lowest detectable concentration of monkeypox virus at which greater than or equal to 95% of all (true positive) replicates test positive.

LoD was evaluated using dilutions of monkeypox virus DNA plasmid in pooled monkeypox virus negative clinical lesion swab VTM specimens. The preliminary LoD study tested 3 target levels at 200, 100, and 50 Copies/mL in replicates of 4 and determined 200 Copies/mL as the lowest concentration that gave positive results 100% of the time. The final LoD was confirmed by testing 21 replicates of the 200 Copies/mL target level, resulting in 100.0% (21/21) detection. The results are summarized in **Table 1**.

Table 1. Limit of Detection Using Monkeypox Virus DNA Plasmid

Target Concentration (Copies/mL)	Number of Replicates		
	Total Valid	Reported Positive	Positive Rate (%)
200	21	21	100.0

LoD was further evaluated using a cultured monkeypox virus (BEI Catalog No. NR-27, Lot 3564851, 2.8 X 10^7 $TCID_{50}$/mL) that was inactivated and diluted in pooled monkeypox virus negative clinical lesion swab VTM specimens. The preliminary LoD study tested 6 target levels at 30, 25, 20, 15, 10 and 5 $TCID_{50}$/mL, each in replicates of 4. The final LoD was confirmed by testing 25 and 30 $TCID_{50}$/mL, each in replicates of 21. The lowest concentration level with observed positive rate ≥ 95% was 30 $TCID_{50}$/mL. Refer to **Table 2**.

Table 2. Limit of Detection Using Cultured Monkeypox Virus

Virus Concentration ($TCID_{50}$/mL)	Number of Replicates		
	Total Valid	Reported Positive	Positive Rate (%)
25	21	18	85.7
30	21	21	100.0

Analytical Specificity – Potentially Interfering Substances

The assay uses a conventional, well-established nucleic acid extraction method; therefore, interference from common endogenous substances is not expected. Interference studies have not been performed for this assay.

Carryover

The carryover rate for Alinity m MPXV was determined by analyzing 361 negative samples processed from alternating positions with high positive samples containing monkeypox virus targeted at 100,000,000 Copies/mL, across a total of 15 runs. Monkeypox virus DNA was not detected in any of the negative samples, resulting in an overall carryover rate of 0.0% (95% CI: 0.0% to 1.1%).

Inclusivity, *in silico* analysis

Inclusivity was demonstrated by analyzing the sequences of the Alinity m MPXV primer and probe sets for homology with 1364 full-length genomic sequences (32 Clade I sequences, 1330 Clade II sequences, and 2 unspecified) of the monkeypox virus isolates available in GISAID EpiPox™ database as of September 12, 2022. None of the sequences have mismatches in the Alinity m MPXV assay target regions. This *in silico* analysis predicts the detection of the analyzed monkeypox virus isolates by Alinity m MPXV assay.

Cross-Reactivity, *in silico* analysis

Related pathogens and high prevalence disease agents that are reasonably likely to be encountered in the clinical specimen (listed in **Table 3**) have been evaluated *in silico* to identify the % homology between the selected primer and probe sequences and the sequence present in the microorganism.

The conclusion of this analysis is that there is limited opportunity for cross-reactivity to allow for false-positive reporting based upon the following:

- Amplification is unlikely due to none or only one of the primers having >80% homology.
- Mismatches in the 3′ end of a primer makes extension unlikely.
- Forward and reverse primer being positioned on the same strand will result in no amplification.
- The distance between the forward and reverse primers >1000 nucleotides makes amplification unlikely.
- Probe is unlikely to bind to target due to genomic analysis of the following microorganisms (refer to **Table 3**).

Table 3. Microorganisms

Acinetobacter calcoaceticus	HPV 18
Acinetobacter lwoffii	Human Genomic DNA
Actinomyces israelii	*Kingella kingae*
Adenovirus type 1	*Klebsiella pneumoniae*
Adenovirus type 7	*Lactobacillus* genus
Bacteroides fragilis	*Listeria monocytogenes*
BK Virus	*Mobiluncus mulieris*
Bordetella pertussis	Molluscum contagiosum virus
Camelpox virus	*Moraxella catarrhalis*
Campylobacter jejuni	*Mycobacterium tuberculosis*
Candida albicans	*Mycoplasma genitalium*
Chlamydophila pneumoniae	*Mycoplasma pneumoniae*
Chlamydia trachomatis serovar D	*Neisseria gonorrhoeae*
Chlamydia trachomatis serovar I	*Neisseria meningitidis*
Clostridium difficile	*Propionibacterium acnes*
Clostridium perfringens	*Proteus mirabilis*
Corynebacterium diphtheriae	*Proteus vulgaris*
Corynebacterium jeikeium	*Pseudomonas aeruginosa*
Cowpox Virus	*Staphylococcus aureus*
Cryptococcus neoformans	*Staphylococcus epidermidis*
Cytomegalovirus	*Staphylococcus saprophyticus*
Ectromelia (mousepox) virus	*Streptococcus agalactiae*
Enterobacter cloacae	*Streptococcus* Group C
Enterococcus faecium	*Streptococcus* Group G
Enterococcus faecalis	*Streptococcus mitis*
Epstein-Barr virus	*Streptococcus mutans*
Escherichia coli	*Streptococcus pneumoniae*
Fusobacterium nucleatum	*Streptococcus pyogenes*
Haemophilus ducreyi	*Streptococcus salivarius*
Haemophilus influenzae	*Treponema pallidum*
Hepatitis B virus	*Trichomonas vaginalis*
Hepatitis C virus	*Trichophyton rubrum*
Herpes Simplex Virus (HSV-1 and HSV-2)	*Toxoplasma gondii*
Herpes Virus 6A strain GS	Vaccinia virus
Herpes Virus 6B strain Z29	Varicella-zoster virus (chickenpox)
HIV-1	Variola virus (smallpox)
HPV 16	

Clinical Performance Evaluation

A clinical evaluation study was performed to evaluate 36 contrived positive clinical samples with the Alinity m MPXV assay. Each contrived positive clinical sample was prepared using a negative lesion swab specimen in VTM, spiked with a monkeypox virus-positive specimen that had been previously inactivated, targeting 2x and 4x LoD. In addition to the contrived positive samples, 35 individual presumed negative lesion swab specimens were tested with the Alinity m MPXV assay.

The results are summarized in **Table 4.** The percent agreement is summarized in **Table 5**. All positive samples were detected (36/36). None of the negative samples were detected (0/35).

Table 4. Clinical Performance Evaluation--Alinity m MPXV Assay

Monkeypox Virus Concentration	Number of Valid Samples	Number Reported Positive
2x LoD	18	18
4x LoD	18	18
Negative	35	0

Table 5. Clinical Performance Evaluation--Agreement		
	Percent Agreement	**95% Score CI**
PPA	100.0% (36/36)	(90.4%, 100.0%)
NPA	100.0% (35/35)	(90.1%, 100.0%)

PPA=Positive Percent Agreement
NPA=Negative Percent Agreement

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KEY TO SYMBOLS

REF	Reference Number
IVD	In Vitro Diagnostic Medical Device
In Vitro Test	In Vitro Test
LOT	Lot Number
AMP TRAY	AMP TRAY
ACT TRAY	ACT TRAY
Rx ONLY	Prescription Only
	Systemic Health Effects
	Warning
	Consult Instructions for Use
	Temperature Limitation
	Use By
	Sufficient for
	Manufacturer

TECHNICAL ASSISTANCE

For technical assistance, call Abbott Technical Services at 1-800-553-7042 (within the US) or +49-6122-580 (outside the US), or visit the Abbott website at www.molecular.abbott.

Abbott Molecular Inc. is the legal manufacturer of the:
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