



Client Communication

Monkeypox Virus PCR Test

Effective August 24, 2022

Pathology Laboratories, Inc. (PathLabs) is announcing a new test offering for Monkeypox Virus PCR, that will be performed at our sister laboratory, Sonic Reference Laboratory (SRL), effective August 24, 2022. This test can be performed on clinical specimens collected during the acute/symptomatic phase of illness from patients suspected of Monkeypox. Further details are disclosed in this document.

Test Name	Test Code	Ordering Recommendation
MONKEYPOX VIRUS PCR	39448	For the detection of non-variola Orthopoxvirus DNA in clinical specimens including Monkeypox virus.

Specimen Requirements

Container Type: Sterile leak-proof screw cap tube preferably with O-ring or gasket seal; Leak-proof sterile screw cap tube; Swabs in viral transport medium (VTM).

Specimen Type: 2 swabs from skin lesions.

Handling Instructions: Collect 2 swabs from each lesion. Swab 2-3 lesions from different parts of the body or lesions with differing appearance. Do NOT collect and submit swabs from more than 3 lesions.

Vigorously swab or brush active lesion with 2 separate sterile synthetic swabs (e.g. Dacron, polyester, or nylon) with a plastic shaft and breakpoint close to the tip.

DO NOT USE COTTON SWABS. Break off or cut the tip of each swab into a sterile tube or tube with 1 mL of VTM. Each swab should be placed in a separate tube. Parafilm may be used to ensure a leak-proof lid if the tube does not have an O-ring or gasket seal. The length of the swabs after breaking or cutting should be no longer than 35mm (1.5 inches).

DO NOT ADD TRANSPORT MEDIA TO SPECIMENS INTENDED TO BE SHIPPED AS DRY SWABS. FREEZE IMMEDIATELY AFTER COLLECTION.

Transport: Frozen

Specimen Stability: Refrigerated: 7 Days; Frozen: 1 Month

Rejection Criteria: Swabs from more than 3 lesions. Single or cotton swabs. Swabs with wooden or metal shafts. Swabs in transport media other than VTM. Out of stability specimens. Ambient specimens.

Methodology: Real-Time Polymerase Chain Reaction (RT-PCR)



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Specimen Requirements (continued)

Analytic Time: 3-4 Days; Tested Monday through Friday

CPT Code: 87593

Compliance Statement: FDA cleared or approved assay.

Orderable Code

Order Code	Reporting Title			
39448	MONKEYPOX VIRUS PCR			
	RESULT	CODE	LOINC	CPT
	394481	MONKEYPOX VIRUS PCR	100434-0	87593
	394482	SPECIMEN TYPE (Ask at Order Entry)	31208-2	
	394485	SOURCE SITE (Ask at Order Entry)	39111-0	
	394483	SYMPTOMATIC (Ask at Order Entry)	95419-8	
	394484	SYMPTOM ONSET DATE (Ask at Order Entry)	11368-8	
	PATIENT DEMOGRAPHICS (Ask at Order Entry)			

Ask at Order Entry - Required Answers

394482 SPECIMEN TYPE (see list of Specimen Types below)

SPECIMEN TYPE	DESCRIPTION
BX	BIOPSY
DL	DRY LESION SWAB
ND	NODULE
NO	NOT SPECIFIED
SC	SCAB SPECIMEN (CRUST)
TO	TOUCH PREP
VF	VESICLE FLUID
VM	SWAB in VTM
WL	WET LESION SWAB

394485 SOURCE SITE (Free Text Description - Examples: Abdomen, Left Hand, Leg, etc.)

394483 SYMPTOMATIC (Entries must be one of the following: **Y**-Yes, **N**-No, **U**-Unknown)

394484 SYMPTOM ONSET DATE (Date Format: mm/dd/yyyy)

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Patient Demographics

(Required for Department of Health Reporting)

1. Address
2. City
3. State
4. Zip
5. Phone

NOTE: RACE AND ETHNICITY ARE NOT REQUIRED. The default answer is UNKNOWN, if not known or not provided.

Bidirectional Interface: For those clients ordering through a **bidirectional interface**, this demographic information is provided in the HL7 PID (*patient identification*) segment.

Manual Requisitions: **Manual requisitions** must show complete patient demographic data as required.

Our online test directory will be updated to reflect this information on August 24, 2022.

This Client Communication will be posted to our website for reference.

Please review the information and make changes as applicable to your practice/facility. If you have any questions, please contact our Client Service Department at 419.255.4601/800.281.8804 or your account executive. Thank you.

NOTE 1: CPT codes are provided for information only, and are based on Pathology Laboratories' current understanding of Medicare rules and carrier instructions and in accordance with the current issue of physicians current procedural terminology (CPT), published by the American Medical Association. Medicare coding may differ from coding used by other third party payers. Questions regarding coding should be confirmed with the payer being billed. Pathology Laboratories cannot accept responsibility for the reimbursement clients may or may not receive based on the procedure codes provided.

NOTE 2: Sonic Reference Laboratory (SRL), an affiliate of Sonic Healthcare USA, Inc., began offering the Monkeypox Virus by PCR test on July 18, 2022. SRL's New Test Announcement is enclosed for reference. Please reference CLINICAL INFORMATION on pages 4-6 of this publication.

Clinical Information:

Monkeypox is a zoonotic disease caused by monkeypox virus, an enveloped double-stranded DNA virus that belongs to the *Orthopoxvirus* genus. Other clinically important viruses of the *Orthopoxvirus* genus are variola virus (the causative agent of smallpox), vaccinia virus (used in the smallpox vaccine), and cowpox virus. Monkeypox was first identified as the causative agent of two pox-like disease outbreaks in monkey colonies housed for research in 1958. Although the exact natural host remains unknown, the monkeypox virus has been isolated from rope squirrels, tree squirrels, Gambian pouched rats, dormice, non-human primates, and other species.

The first case of human monkeypox was identified in 1970 in the Democratic Republic of the Congo (DRC) in a 9 month-old child. Historically, monkeypox outbreaks have been mostly confined to the endemic area, specifically the Congo Basin/DRC. However, the virus has been in circulation in several countries of Western and Central Africa. The rise in monkeypox outbreaks is believed to be due to eradication of smallpox and cessation of mass smallpox vaccination, which offered cross-protective immunity to *orthopoxviruses*. This resulted in a significantly increased population of those immunologically naïve to *orthopoxviruses*. Disease outside of the endemic region of Western and Central Africa was rare and mostly linked to international travel to the endemic region, or through imported animals. The first significant outbreak of human monkeypox outside of the endemic region occurred in the Midwestern United States in 2003 and was associated with imported African rodents. The first case associated with the current 2022 outbreak of monkeypox in non-endemic countries was confirmed in the United Kingdom on May 6 in an individual who had travelled to Nigeria. Following confirmation of the first US case on May 17, 2022, 929 confirmed cases have been reported in the US (as of July 12, 2022). Close to 11,000 cases of monkeypox have been confirmed globally in at least 59 non-endemic countries (Figure. 1).

Sequencing and phylogenetic analysis of isolates from multiple outbreaks demonstrates two geographically separated genetic clades of monkeypox virus, specifically the West African and the Central African clade (also known as the Congo Basin clade). The West African clade is less virulent with a case fatality rate (CFR) of <1% and lacks a number of genes in comparison to the Central African strain. In contrast, the Central African/Congo Basin clade is considered more virulent and has a CFR up to 11%. Monkeypox virus sequences from the ongoing 2022 outbreak, including those from the US, are similar to the sequences of the strain that caused the 2017-2018 Nigeria outbreak, raising the possibility that the 2022 outbreak resulted from importation(s) of virus from an endemic country.

Animal-to-human transmission of monkeypox virus occurs through direct contact with the blood, bodily fluids, or cutaneous or mucosal lesions of infected animals. Human-to-human transmission typically occurs through contact with infectious respiratory secretions, skin lesions of an infected person or recently contaminated objects. Most cases in the current outbreak have been identified in men who have sex with men, leading to the possibility that monkeypox can be transmitted through sexual transmission routes. Transmission can also occur through contact with materials or fomites (e.g. clothing or linens) that have been contaminated with bodily infectious materials. Transmission has also been reported via droplet respiratory particles, via the placenta from mother to fetus or via close contact during and after childbirth.

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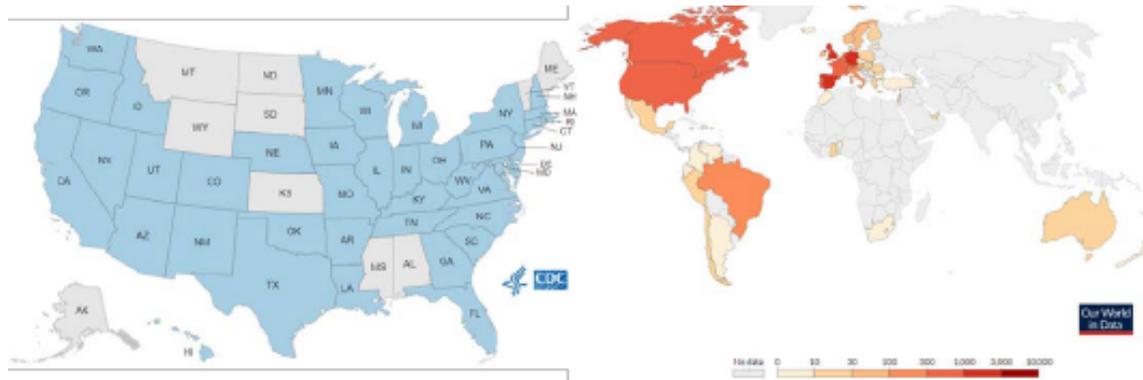


Figure 1. Cumulative confirmed cases of monkeypox virus in the US and non-endemic countries as of July 12, 2022.

The incubation period of monkeypox can be 5 to 21 days but typically ranges from 6 to 13 days. The disease manifestation can be divided into two periods. The initial prodromal/invasion period is characterized by fever, intense headache, lymphadenopathy, back pain, myalgia and intense asthenia within 5 days of infection. Whereas the rash/skin eruption usually occurs within 1 to 3 days of the appearance of fever and continues for a period of 2 to 3 weeks. The rashes are usually concentrated on the face and extremities (palms of the hands and soles of the feet). Other affected sites may include oral mucous membranes, genitalia, and conjunctivae, as well as the cornea. The rash progresses successively from macules to papules, vesicles, pustules, and crusts that dry up and fall off. The number of lesions can be highly variable, from a few to several thousand. In severe cases, lesions can coalesce until large sections of skin slough off. The disease is generally self-limiting and the symptoms last from 2 to 4 weeks. An individual remains infectious from the onset of illness until all lesions have scabbed over and re-epithelialization has occurred. Complications of monkeypox can include secondary infections, bronchopneumonia, sepsis, encephalitis, and infection of the cornea with ensuing loss of vision. Severe cases and complication may occur in children and immunocompromised hosts. Asymptomatic infections have been documented but the frequency/prevalence of asymptomatic infections is unknown.

Laboratory diagnosis of monkeypox in suspected individuals can be established by detection of the virus or the immunoglobulin (Ig)M antibodies. Isolation of monkeypox virus in culture from a clinical specimen can be performed but the methodology is cumbersome, slow and requires BSL-3 containment facilities. The CDC instructs routine clinical laboratories not to perform culture-based testing for monkeypox virus. Serologic testing may be particularly helpful if direct viral testing cannot be performed. Molecular detection of monkeypox virus DNA in lesion samples by real-time PCR testing is the preferred and rapid method for viral testing.

SRL's Monkeypox Virus by PCR test methodology is real-time PCR utilizing the CDC Non-variola Orthopoxvirus Real-time PCR Primer and Probe Set. The assay detects non-variola Orthopoxvirus DNA in clinical specimens. Although this assay does not differentiate monkeypox virus from other Orthopoxvirus including vaccinia, cowpox, camelpox, ectromelia or gerbilpox virus, a positive result with this assay in the United States is most likely due to monkeypox virus; however, potential exposure to other Orthopoxviruses should be considered. The test results are intended to be utilized in conjunction with clinical presentation, epidemiological data, other diagnostic test results, and vaccination and exposure history. As monkeypox virus infections can present with rashes/lesions similar to that of varicella zoster virus, herpes simplex virus, and other sexually transmitted infections, consider testing for other pathogens as clinically indicated.

Extensive additional information on monkeypox can be found at the CDC's website.

References

1. US Monkeypox outbreak 2022. CDC's 2022 U.S. Map & Case Count. <https://www.cdc.gov/poxvirus/monkeypox/response/2022/us-map.html>
2. Global Monkeypox outbreak 2022. CDC. <https://www.cdc.gov/poxvirus/monkeypox/response/2022/world-map.html>
3. KD Reed, et al., The detection of monkeypox in humans in the Western Hemisphere. N Engl J Med. 2004. 350, 342- 350. <https://pubmed.ncbi.nlm.nih.gov/14736926/>
4. Sklenovský N, Van Ranst M. Emergence of Monkeypox as the Most Important Orthopoxvirus Infection in Humans. Front Public Health. 2018 Sep 4;6:241. <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6131633/>
5. Monkeypox fact sheet. World Health Organization. <https://www.who.int/news-room/factsheets/detail/monkeypox>
6. Laboratory Procedures and Biosafety Guidelines. CDC. <https://www.cdc.gov/poxvirus/monkeypox/labpersonnel/lab-procedures.html>
7. Collection, Storage, and Shipment of Specimens for Monkeypox Diagnosis. CDC. <https://www.cdc.gov/poxvirus/monkeypox/clinicians/prep-collection-specimens.html>