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Diagnostic considerations in suspected cases of monkeypox

To the Editor: On July 23, 2022, monkeypox was declared a public health emergency of international concern by the World Health Organization.[1] As the case numbers rise exponentially in nonendemic regions, the focus is on strategies to mitigate the spread of the disease. These efforts include establishing a timely diagnosis, early identification of close contacts, and vaccination measures. We aim to summarize the key screening strategies and diagnostic protocols for monkeypox, which could serve as a reference for clinicians when evaluating a "suspected case" of monkeypox.

The current incidence of monkeypox in the United States is 5.2 cases per 1,00,000 population.[2] As per the Center for Disease Control case definitions, if a patient meets any one epidemiologic criterion and has a characteristic rash without an alternative explanation, then he/she is labeled as "suspected monkeypox" (Table I).[2,3] The characteristic rash of monkeypox evolves from macules to umbilicated vesiculopustules in a centrifugal distribution. Clinicians commonly see patients in the vesiculopustular phase when they present with a fever and vesiculopustular exanthem, which may be painful and is often accompanied by lymphadenopathy. The vesiculopustules of monkeypox are firm, difficult to deroof, and lack fluid contents after

Table I. Clinicoepidemiologic characteristics of a suspected case of monkeypox

Suspected case*
- New characteristic rash **OR**
- Meets one of the epidemiologic criteria and has a high clinical suspicion for monkeypox

Exclusion criteria
- An alternative diagnosis (ie, secondary syphilis, herpes, and varicella-zoster) can fully explain the illness **OR**
- An individual with symptoms consistent with monkeypox does not develop a rash within 5 days of illness onset **OR**
- A case where high-quality specimens do not demonstrate the presence of *Orthopoxvirus* or *Monkeypox virus* or antibodies to orthopoxvirus

Exposures
- Incubation period 5-21 days

Within 21 days of illness onset:
- Reports having contact with a person or people with a similar appearing rash or who received a diagnosis of confirmed or probable monkeypox **OR**
- Had close or intimate in-person contact with individuals in a social network experiencing monkeypox activity, this includes men who have sex with men **OR**
- Traveled outside the United States to a country with confirmed cases of monkeypox or where the *Monkeypox virus* is endemic **OR**
- Had contact with a dead or live wild animal or exotic pet that is an African endemic species or used a product derived from such animals (eg, game meat, creams, lotions, powders, etc.)

Signs and symptoms
- Prodrome 1-5 days before the onset of rash
- Fever, chills, malaise, myalgia, arthralgia, headache
- Lymphadenopathy
- Nonproductive cough
- Pharyngitis, odynophagia, epiglottitis,
- Rectal pain, proctitis, tenesmus, diarrhea

Cutaneous findings

Exanthem
- Often starts on the face, anogenital regions, palms, and soles
- Evolves from macules to papules to vesculopustules, may be painful
- Vesiculopustules are umbilicated and progress to crusted lesions, which can scar
- Prodrome to scabs ~2 weeks
- Usually <20 lesions, often in different stages; however, in one given anatomic site lesions are in the same stage

Enanthem
- Anogenital ulcers (may be the first and only presentation)
- Oral or tonsillar lesions
- Nasal or ocular lesions

*Per the 2022 Case Definitions issued by the Centers for Disease Control and Prevention.

Table II. Monkeypox diagnostic procedures, protocol, and special considerations

Specimen collection
- Dry swabs* (regular, commercially available viral or bacterial swabs can be used)
- Macules, papules, vesiculopustules-vigorously swab the surface of the lesion
- Two swabs from each lesion, preferably from different anatomic locations on the body, or from lesions that differ in appearance.
- Place each swab or crust in a separate container, such as a screw-capped tube with an O-ring or another sterile leak-proof container (eg, sterile urine cup)
- Do not use cotton swabs.
- Crusts†-use a forceps or a blunt-tipped sterile instrument to remove all or a piece of the crust that is at least 4 mm × 4 mm

Special considerations
- Deroofing is not essential; lesions do not have to be fluid filled
- Specimens should be double bagged before dropping off
- Pharyngitis-naso-oro-pharyngeal swabs
- Rectal pain/proctitis-Anorectal swabs can be performed
- PPE should be used. This includes gloves, gowns, eye protection (face shields or eye goggles), masks with N95 filters or higher

Diagnostic testing

PCR or next-generation sequencing of a clinical specimen
- Level 1 PCR test—Regional LRN. Sample tested for nonvariola orthopox DNA. Average turnaround time is 3-5 days. If present, label "Probable" case.
- Level 2 PCR test—performed sequentially if results are positive for nonvariola orthopox DNA. Includes further characterization testing at CDC with Monkeypox virus-specific real-time PCR and genetic sequencing. If isolated, label "Confirmed" case.

Testing sites

Health care provider to contact the state health department to identify regional LRN.

For example, New York specimens can be tested at the state's Wadsworth Center, the New York City Department of Health, and at private laboratories, including Quest, LabCorp, Mayo Clinic, Aegis Sciences Corporation, Sonic Healthcare, and UR Medicine Lab.

Tests for research purposes

Not currently available for routine diagnostic purposes. Consult CDC before submission.
- Antiorthopoxvirus IgM antibody during 4-56 days after rash onset
- Isolation of *Monkeypox virus* in culture

Other tests‡
- Histopathology-nonspecific. Early lesions show ballooning degeneration of keratinocytes and spongiosis; later show full thickness necrosis of acanthotic epidermis. Dense inflammatory infiltrate. Multinucleated syncytial keratinocytes, representing viral cytopathic effect can be seen.
- Tzanck smears-Not indicated routinely. Can be helpful to diagnose mimickers like herpes virus infection (multinucleate keratinocytes), and molluscum contagiosum (basophilic intracytoplasmic Henderson Peterson bodies).
- Testing for sexually transmitted infections
  - o Syphilis and HIV: Nontreponemal tests (RPR/VDRL), fourth generation ELISA for HIV
  - o Gonorrhea and chlamydia: NAAT from lesional swabs or urine samples
  - o HSV: Lesional swabs for PCR analysis or HSV-1 and HSV-2 IgM (acute phase) and IgG (convalescent phase)

CDC, Centers for Disease Control and Prevention; *ELISA*, enzyme-linked immunosorbent assay; *HIV*, human immunodeficiency virus; *HSV*, herpes simplex virus; *IgM*, immunoglobulin M; *LRN*, laboratory response network; *NAAT*, nucleic acid amplification test; *PCR*, polymerase chain reaction; *PPE*, personal protective equipment; *RPR*, rapid plasma region; *VDRL*, venereal disease research laboratory test; *VTM*, viral transport media.

*Some laboratories are now accepting swabs in viral transport medium. Bacterial swabs are sturdier than viral swabs.

†Crusts may not be accepted by all laboratories as an approved specimen type.

‡Not indicated routinely to determine a diagnosis of monkeypox but may help rule out mimickers or coinfections.

deroofing—more appropriately referred to as "pseudovesiculopustules."[4]

As shown in Table II, we highlight the specimen collection process in monkeypox, which differs from other morphologically similar vesiculopustular eruptions. For example, in monkeypox, swabs can be collected from intact vesiculopustules and scabs/crusts.[2] This is in contrast to herpetic exanthems, where specimens should be obtained from deroofed fluid-filled vesiculopustules, followed by vigorous

swabbing of the base. Two types of polymerase chain reaction tests, nonvariola orthopox virus DNA and Monkeypox virus DNA isolation, are used sequentially to confirm the diagnosis. The US Department of Health drastically increased the testing capacity by authorizing commercial laboratories to perform monkeypox testing. Testing should be performed at biosafety level 2 facilities with vaccinated personnel. A skin biopsy is not indicated for diagnosis because findings are non-specific. The current standard of care does not include alternative tests like antiorthopoxvirus immunoglobulin M antibodies, immunohistochemical staining, electron microscopy, or viral cultures.[2]

Besides the exanthem, monkeypox can lead to an enanthem, which can be challenging to diagnose.[5] Involvement of oropharyngeal mucosa leads to tongue ulcers and pharyngitis. Anogenital mucosal involvement may be the first and only presenting feature of the disease. A large proportion of cases present with isolated genital ulcers, rectal pain, proctitis, tenesmus, or diarrhea.[5] In such cases, besides sampling these sites, clinicians must consider additional testing for other sexually transmitted infections, which can mimic, or co-occur with monkeypox (Table II).

In the current epidemic, dermatologists should have a high index of suspicion, and a low threshold for swabbing suspicious lesions. A comprehensive awareness of the clinical features of monkeypox and its mimickers, coupled with knowledge of the diagnostic protocols, will equip clinicians to determine a diagnosis promptly and accurately. It will also help reduce "false alarm" diagnoses, which may lead to increased health care costs and negative psychosocial impact on patients stigmatized and isolated by the diagnosis.

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Conflicts of interest

None disclosed.

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