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Introduction

This procedure provides general guidance on how to handle specimens from patients with suspect or confirmed monkeypox.

Specimen Collection and Storage

1. Recommended specimens varying on the patient’s presenting signs/symptoms.
   - **Throat swab/NP swab** and **serum** may be requested at the prodromal stage along with **lesion fluid/crust/scab, lesion swab** at the skin rash stage – all for **Monkeypox Virus Detection**.

2. Please refer the list below for the acceptable specimen types:

<table>
<thead>
<tr>
<th>Specimen Type</th>
<th>Minimum Volume</th>
<th>Comment</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td>≥0.5mL</td>
<td>• Red top&lt;br&gt; • Serum separator tube (SST)&lt;br&gt; • Centrifugation of serum tubes is not needed and should be avoided</td>
</tr>
<tr>
<td>Lesion fluid, crust material or scab</td>
<td>Not Applicable</td>
<td>• Sterile 1.5-5.0mL tube/container with or without transport media</td>
</tr>
<tr>
<td>Swab of lesion</td>
<td>Not Applicable</td>
<td></td>
</tr>
<tr>
<td>Nasopharyngeal and/or oropharyngeal swab</td>
<td>Not Applicable</td>
<td></td>
</tr>
<tr>
<td>Cerebrospinal fluid (CSF) (if encephalitic)</td>
<td>≥0.5mL</td>
<td></td>
</tr>
<tr>
<td>Urine (if severely ill)</td>
<td>50mL</td>
<td></td>
</tr>
<tr>
<td>Tissues</td>
<td>Not Applicable</td>
<td></td>
</tr>
<tr>
<td>Formalin-fixed, or paraffin embedded tissues</td>
<td>Entire blocks, or four to six 10 micron block sections</td>
<td>Sterile container (clearly label if formalin fixed)</td>
</tr>
</tbody>
</table>

3. All specimens will be cleaned and placed in specimen transport bags then deliver to the lab
   - **NOTE:** the pneumatic tube may be used, however skin samples are expected to arrive in Zip N’ Fold bags.
Tests Performed in SHS/UHN Department of Microbiology

The tests listed below are performed in SHS/UHN Department of Microbiology with the initial specimen processing to be completed following safety precautions as stated in specimen processing procedure below.

a. Routine bacteriology
b. Routine mycology
c. HSV/VZV on Aries
d. COVID-19 on Aries/GeneXpert
e. Influenza/RSV on Aries/GeneXpert
f. C. difficile toxin B detection on Aries/GeneXpert
g. Serology testing on Alinity
h. Virology testing on cobas6800
i. PCR testing on easyMag-Biorad platform

Note: Other tests that impact patient care may be considered upon consultation with the microbiologist-on-call

Reagents/Materials/Media

1. Accel PREVention Wipes
2. Necessary suppliers for the tests performed in-house
3. Personal protective equipment:
   - Fit-tested N95 (choose fit tested model and size)
   - Disposable face shield
   - Disposable water resistant gown
   - Disposable nitrile gloves

Specimen Processing

1. Specimens should be received as soon as they are arrive in the laboratory. 
   NOTE: centrifugation of serum tubes is not needed and should be avoided.

2. Don PPE while handling specimens
3. Disinfect the bag and specimens with AccelPREVention Wipes
NOTE: Page microbiologist-on-call through locating for any discrepancies between samples and requisition

4. Receive all orders in the LIS system and add in the RG3 alert “MPX-1”.
5. print the LIS labels including the media labels for the tests that will be performed in-house
6. Label all specimens with correct LIS label
7. Set up all in-house tests in BSC according to the appropriate protocol for the indicated test
8. Disinfect the BSC using Accel PREVention Wipes
9. Proceed with in-house tests as per usual protocols in the BSC
10. Discard all wastes from the samples under monkeypox investigation in biohazard bin prior to sending out for incineration.
11. Use the following test codes for monkeypox virus detection:

   - MOPXX – Monkeypox Virus Detection (Other)
   - MOPXB – Monkeypox Virus Detection (Blood)

12. Order Virology Refer Out using keypad for the 1st additional send-out PCR test (e.g. Enterovirus)
13. Order Virology PCR using keypad for the 2nd additional sent-out PCR test
14. Place LIS sticker in the template below when sending out samples for Monkeypox
15. Keep the log sheet in the binder on the Virology Accessioning Bench

<table>
<thead>
<tr>
<th>Monkeypox sendout</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>LIS#</strong></td>
<td><strong>Date sent out</strong></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Specimen Transportation for Monkeypox Virus Detection

I. Shipping Condition

<table>
<thead>
<tr>
<th>Sample Type</th>
<th>Shipping Condition (Time between collection and shipping)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Within 72 hours (≤72hrs)</td>
</tr>
<tr>
<td>Lesion fluid, crust material or scab</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Swab of lesion</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Nasopharyngeal and/or throat swab</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Serum</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Cerebrospinal fluid (CSF)</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Urine</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Tissues</td>
<td>2-8°C (Ice Pack)</td>
</tr>
<tr>
<td>Formalin-fixed, or paraffin embedded tissues</td>
<td>Room Temperature</td>
</tr>
</tbody>
</table>

II. Shipping Documents/Transportation Preparation

1. Print PHOL requisition using MIC10 format from LIS
   The following information must be provided on PHOL requisition
   1.1. Patient identifiers
   1.2. Number of specimens being sent
   1.3. Indicate Monkeypox Virus Detection as the requested test
   1.4. Include clinical information from order entry. (Symptoms and onset, Risk factors/exposures, travel history, VZV vaccination history etc)

2. Package monkeypox test samples in a separate CAT B UN3373 compliant shipping box, labelled with pink TU0886 sticker.

3. Arrange for transportation to PHOL using Category B shipment protocols (as directed by Temporary Certificate TU0886).
   2.2. Ship specimens at 6am each weekday to PHOL
   2.3. When ready to ship, contact KJV for transportation

4. Email ICP of submitting hospital that the specimen was received and has been sent to PHOL for Monkeypox Virus Detection with the associated specimen LIS number.

UNIVERSITY HEALTH NETWORK/MOUNT SINAI HOSPITAL, DEPARTMENT OF MICROBIOLOGY

NOTE: This document is Uncontrolled When Printed.
Any documents appearing in paper form that do not state “CONTROLLED COPY” in red print are not controlled and should be checked against the document (titled as above) on the server prior to use.
Phrases for deferring non-monkeypox tests

For tests that can be deferred:
“This test has been deferred for biosafety reasons until monkeypox is ruled out. Please contact the microbiologist-on-call with any questions.”

For tests that can’t be deferred and need to be cancelled:
“This test was CANCELLED for biosafety reasons given that monkeypox is being ruled out. Please contact the microbiologist-on-call with any questions.”

For tests that are cancelled after positive monkeypox results:
“This test was CANCELLED for biosafety reasons given positive monkeypox test results. Please contact the microbiologist-on-call with any questions.”

For tests set up late due to initial deferral:
“Testing was delayed until monkeypox was ruled out which may reduce the sensitivity of this test. Please take this into consideration when interpreting this result. Please contact the microbiologist-on-call with any questions.”
### Resulting from PHOL Report

1. **Negative Result** – Result in the **VSEND** test comment:
   1.1. Press “**F8**” to open Test Comment Window
   1.2. Select **Y >MOP** from VSEC-1 of 1 window
   1.3. Select **1 )-MOP**
   1.4. **Finalize** the report
2. **Invalid Result** – Result in the VSEND test comment:
   
   2.1. Press “F8” to open Test Comment Window
   2.2. Select *Y >MOP* from VSEC-1 of 1 window
   2.3. Select 4 }MINV
   2.4. **Finalize** the report

   ![VSEND Test Comment Window]

   ![MOP Test Comment Window]

3. **Positive Result** – Result in the Isolate window.
   
   3.1. Add isolate *75monkey*
   3.2. Press “F8” to open Isolate Comment Window
   3.3. Isolate comment \Dna automatically appends
   3.4. Below this comment add \Mpx+
   3.5. Type in appropriate results as well as PHOL#
   3.6. **Interim** the report

   ![Isolate Comment Window]
4. **Indeterminate Result** - Result in VSEND test comment.

4.1. Press “F8” to open Test Comment Window

4.2. Select Y >MOP from VSEC-1 of 1 window

4.3. Select 3 }MIND

4.4. **Interim** the report

4.5. **Ensure** to set significant status with Red check mark in LIS by right clicking mouse
Result Communication

1. Communicate any verbal results (positive/negative/invalid/indeterminate) according to the isolate notification table in the internal manual.

2. Result in LIS only when hard copies are received from PHOL.

3. Remove RG3 “MPX-1” alert in the LIS if ALL monkeypox results are NEGATIVE; verbal reports are sufficient to remove the RG3 alert. Do not remove alert if there are monkeypox results still pending.

4. All results will be notified to the Medical Officer of Health by Public Health Ontario.

Troubleshooting

1. Follow up any monkeypox orders that have not been reported after 48 hours of receipt in PHOL.

   1.1. Call PHOL customer service if no monkeypox report available in MSH Microbiology Lab
   1.2. If PHOL customer service does not have the results, make sure to ask them to investigate the delay with the PHOL lab technologist.
2. Page microbiologist-on-call through locating for any other situation not included in this procedure

Reference

1. PHOL: https://www.publichealthontario.ca/en/Laboratory-Services/Test-Information-Index/Monkeypox-Virus
2. NML: https://cnphi.canada.ca/gts/reference-diagnostic-test/5030?labId=1021
3. CDC: https://www.cdc.gov/poxvirus/monkeypox/lab-personnel/lab-procedures.html
Record of Edited Revisions

Manual Section Name: Standard Operation Procedure Template

<table>
<thead>
<tr>
<th>Page Number / Item</th>
<th>Date of Revision</th>
<th>Signature of Approval</th>
</tr>
</thead>
<tbody>
<tr>
<td>New</td>
<td>May 27, 2022</td>
<td>Dr T Mazzulli</td>
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</table>

Full document review included in all updates. Bi-annual review conducted when no revision had been made within 2 years.

<table>
<thead>
<tr>
<th>Page Number / Item</th>
<th>Date of Revision</th>
<th>Edited by:</th>
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<tbody>
<tr>
<td>Added in-house testing platform</td>
<td>June 01, 2022</td>
<td>Qin Liu</td>
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<tr>
<td>Ensure to set significant status for positive result</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Updated Transport Canada requirement TU0886</td>
<td>June 8, 2022</td>
<td>Wayne Chiu</td>
</tr>
<tr>
<td>Updated PHOL process</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Updated BSC information, ensure to use BSC with PPE (no need for 2.5)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Updated disposal information, follow routine RG3/4 procedure and send for incineration (no need to autoclave inhouse)</td>
<td></td>
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<tr>
<td>Removed responsibility of microbiologist to notify ID</td>
<td></td>
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<tr>
<td>Update Resulting from PHOL report</td>
<td>June 10, 2022</td>
<td>Qin Liu</td>
</tr>
<tr>
<td>Update result communication:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Only result in LIS when hard copies received from PHOL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Add the updated result from NML report</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Add the trouble shooting for any orders with no results after 48 hours receipt in PHOL</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Added monkeypox test codes</td>
<td>June 17, 2022</td>
<td>Qin Liu</td>
</tr>
<tr>
<td>Deleted the actual resulting words</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Multiple updates re microbiologist approval, transport, pneumatic tube ok, sendout frequency</td>
<td>June 23, 2022</td>
<td>Wayne Chiu</td>
</tr>
<tr>
<td>Specified need to package monkeypox in separate CAT B shipping box</td>
<td>June 28, 2022</td>
<td>Wayne Chiu</td>
</tr>
<tr>
<td>Removed NML test/report sections</td>
<td>June 30, 2022</td>
<td>Wayne Chiu</td>
</tr>
<tr>
<td>Updated positive report resulting and isolate code</td>
<td>July 5, 2022</td>
<td>Wayne Chiu</td>
</tr>
<tr>
<td>Section:</td>
<td>Bacteriology Procedures</td>
<td>Subject Title: Monkeypox Procedure</td>
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