

Influenza Virus A and B – Qualitative, Real-time PCR

Use

Detection and characterization of influenza virus types A (subtypes H1, pdm09, H3, H5, H7), or B in patients with influenza-like symptoms. This test is used primarily to define influenza A subtypes of previously determined influenza A-positive or influenza A-untypeable patient samples.

Note: *Use the Influenza A/B, SARS-CoV-2, and Respiratory Syncytial Viruses – Qualitative, Real-time PCR with Reflex to Influenza Virus A Subtyping – Qualitative, Real-time PCR test if the patient has not been tested for influenza, SARS-CoV-2 or RSV or influenza A/B testing needs to be confirmed.*

OSDH Infectious Disease Services pre-approval is required for submission of specimens from patients with suspected influenza A/H5 or Eurasian H7 to verify the patient meets clinical case criteria, including travel history to an area of novel influenza circulation. Call 24/7/365 for telephone consultation at 405-426-8710.

Clinical Significance

Influenza virus types A, B, and C are RNA viruses that cause acute respiratory disease with associated fever, shivering, chills, headache, malaise, dry cough, loss of appetite, body aches and nausea. Severe cases are associated with prostration, hemorrhagic bronchitis, pneumonia and occasionally death, especially in the young and elderly. Most human infections are caused by influenza types A and B. Current subtypes of influenza A viruses that routinely circulate in people include A(H1N1), A(H3N2), B(Victoria) and B(Yamagata). Most influenza-like infections are not caused by influenza but by other viruses (e.g., rhinoviruses and respiratory syncytial virus, adenoviruses, and parainfluenza viruses).

Further background information, fact sheets, statistics and educational resources may be found at the OSDH Infectious Disease Services [website](#).

Methodology

The Human Influenza Virus Real-time Reverse Transcriptase-PCR Diagnostic Panel was developed by the Centers for Disease Control and Prevention (CDC) to diagnose human infections with seasonal influenza viruses and novel influenza A viruses.

Specimen Type

- Routine samples: Nasopharyngeal (NP) swab
- Suspect influenza A/H5 or A/H7: *(requires OSDH Infectious Disease Service pre-approval prior to submission)*
 - NP swab, or
 - Nasal swab combined with oropharyngeal swab (2 separate swabs placed in one viral transport vial), or
 - If a person has conjunctivitis, NP swab and conjunctival swab (2 separate swabs placed in separate viral transport vials).

Minimum Volume/Size

1 or 2 swabs (see above) in tube(s) containing a suitable viral transport medium; examples include Remel M4RT, M4, or M5, BD Universal Viral Transport (UVT[®]) Medium, and Copan Universal Transport Medium (UTM[®]).

Collection Instructions

Respiratory virus detection depends on the collection of high-quality specimens, their rapid transport to the testing laboratory and appropriate storage before testing. Training in specimen collection is highly recommended due to the importance of specimen quality. Specimens should be collected using standard procedures of the submitting site. Follow storage and collection instructions applicable to the collection kit/medium used. Additional guidance is contained in the links below.

- See [Guidance for Collection of Nasopharyngeal Swab](#)
- See [Guidance for Collection of Oropharyngeal Swab](#)
- See [Guidance for Collection of Nasal Swab](#)
- See [Guidance for Collection of Conjunctival Swab](#)

Common Causes for Rejection

- Incorrect collection device (cotton, wooden or calcium alginate swab)
- Transport media other than those indicated
- Received > 72 hours from time of collection and not frozen
- Swab without transport medium
- Specimens missing swabs
- Specimens that have leaked
- Specimen at ambient temperature

Shipping

- **Refrigerated** (2–8°C), use frozen cold packs: for fresh specimen(s) that will arrive at OSDH PHL within 72 hours of collection.
- **Frozen**, use dry ice: for specimen(s) that will **not** arrive at the OSDH PHL within 72 hours of collection.

Samples may be kept frozen (at least -20°C but -70°C or colder is preferred) at the collection site and submitted to the OSDH PHL if courier delivery cannot be arranged for same or next day delivery. If site is unable to provide dry ice for shipment, please call the OSDH PHL at 405-564-7750 at least 24 hours ahead of scheduled submission to request dry ice transport.

Turn-around Time

Within 5 working days of receipt

Specimens in which variant or potential novel influenza viruses are detected by the Human Influenza Virus RT-PCR Diagnostic Panel may be issued a preliminary report, pending further characterization by the CDC.

Reference Range

Influenza Not Detected

Reportable Results

The OSDH PHL does not have the ability to subtype all strains of influenza virus. Also, some strain results require confirmation by the CDC before they can be reported. However, specimens must meet specific quality criteria to be acceptable for testing at the CDC. If acceptable, the OSDH PHL report will indicate “Subtype: Undetermined, referred to CDC for subtyping” and a CDC report will follow. If unacceptable by CDC, the OSDH PHL report will indicate “Subtype: Undetermined” as the final report.

- Influenza Not Detected
- Influenza Virus A Detected, Subtype: H1 2009 pandemic strain
- Influenza Virus A Detected, Subtype: H3 strain
- Influenza Virus A Detected, Subtype: H1 2009 pandemic strain; possible coinfection or recent live attenuated influenza virus vaccination
- Influenza Virus A Detected, Subtype: H3 strain; possible co-infection or recent live attenuated influenza virus vaccination
- Influenza Virus A Detected, Subtype: Undetermined
- Influenza Virus A Detected, Subtype: Undetermined, referred to CDC for subtyping
- Presumptive Positive for Influenza A/H3N2 variant; referred to CDC for confirmation

- Influenza Virus B Detected
- Influenza Virus B Detected, possible co-infection or recent live attenuated influenza virus vaccination
- Indeterminate: Potential PCR inhibitor or poor quality of sample; recommend recollection and submission of fresh sample, if clinically appropriate

Note: *The OSDH PHL does not have the ability to subtype all strains of influenza A virus. Also, some strain results require confirmation by the CDC before they can be reported. Specimens must meet specific quality criteria to be acceptable for testing by the CDC; if the specimen is acceptable, the OSDH PHL report will indicate "Subtype: Undetermined, referred to CDC for subtyping" and a CDC report will follow, whereas if unacceptable by CDC, the OSDH PHL report will indicate "Subtype: Undetermined" as a final report.*

Interpretation

- Failure to detect RNA for any of the targeted viruses suggests an absence of viruses in the sample; however, negative results do not preclude infection with targeted or other viruses and should not be used as the sole basis for treatment or other patient management decisions.
- Detection of RNA for any of the targeted viruses suggests the presence of targeted virus in the sample; however, positive results do not imply infectivity or rule out other viral or bacterial co-infections and should not be used as the sole basis for treatment or other patient management decisions.
- An indeterminate or inconclusive test result is likely due to a PCR inhibitor or poor-quality specimen.

Limitations/Interferences

- As with any molecular test, mutations within targeted sequences could affect primer and/or probe binding resulting in failure to detect the presence of target viruses or newly emerging variants.
- The performance characteristics of the CDC Human Influenza Virus RT-PCR Diagnostic Panel may vary depending on influenza A variants circulating and their prevalence, which are expected to change over time.
- Viral nucleic acid may persist *in vivo*, independent of virus infectivity. Detection of analyte target(s) does not imply that the corresponding virus(es) are infectious or are the causative agents for clinical symptoms.
- Erroneous test results might occur from improper specimen collection; failure to follow the recommended sample collection, handling, and storage procedures; technical error; or sample mix-up.
- Individuals immunized with live attenuated influenza nasal spray vaccine may be positive for one or more influenza virus targets for several days post-vaccination; vaccination history should be considered when interpreting positive test results, especially early in the respiratory virus season.

CPT Code

87502, 87503 (x3)

Notes

This test is approved for *in vitro* diagnostic use by the U.S. Food and Drug Administration.