Washington State Department of HEALTH

DOH 301-018

Influenza Virus Testing at the Washington State Public Health Laboratories (WAPHL) Including Novel Influenza and Fatal Influenza

April 16, 2025

The WAPHL performs influenza virus testing and subtyping. Results are used to monitor state influenza activity. In special situations, testing may be done to determine if novel influenza infection is occurring in humans. Testing and subtyping are performed using real-time reverse transcriptase polymerase chain reaction (RT-PCR) assays developed by the CDC. A subset of positive samples are sent to CDC for further characterization every 2 weeks.

What influenza testing can be performed at WAPHL?

After approval from the local health jurisdiction, WAPHL will perform influenza testing and subtyping on specimens from:

1. Deceased patients suspected to have influenza. If not screened for influenza specimens will be tested with BioFire Respiratory panel. If positive, genotyping will be performed.
2. Patients with suspected novel influenza virus infection, such as infection with influenza A (H3N2v), (H5N1) or (H7N9) virus. **NOTE**: If novel influenza A virus infection is suspected, specimens should be collected using appropriate infection control precautions and sent IMMEDIATELY to WAPHL.
3. Patients associated with outbreaks.
4. Persons with exposure to avian influenza infected birds or animals.

NEW SUBTYPING RECOMMENDATION: In accordance with the CDC HAN released in January 2025, WA DOH recommends that all patients with suspected influenza are tested within 24 hours of hospital admission and that laboratories expedite seasonal influenza subtyping for all influenza A positive patients, especially ICU patients and patients with highly pathogenic avian influenza (HPAI) epidemiologic risk factors. See figure below.

- If hospitals cannot subtype in-house, send to a commercial laboratory for subtyping, or are challenged with capacity/cost, specimens can be sent directly to PHL.
- Unlike the typical submission process to PHL, all subtyping specimens can be sent to PHL WITHOUT notification to LHJ unless, 1. Specimen is unsubtypeable or 2. Specimen has epi-risk factors or suspect for HPAI. See below for more information.

After approval from the local health jurisdiction, CDC can perform antiviral resistance testing for infection control purposes on specimens from:

1. Patients who develop laboratory-confirmed influenza while taking antiviral prophylaxis.
2. Severely immunocompromised patients with prolonged excretion of influenza virus despite antiviral treatment.
3. Patients in intensive care units with prolonged excretion of influenza virus despite antiviral treatment.

Specimen Collection and Allowable Specimen Types

***The following specimen types are preferred for seasonal influenza testing at WAPHL*:**

- Nasopharyngeal swab, nasal aspirate/wash, or dual nasopharyngeal/throat swab.
- The following specimen types are also acceptable for influenza testing at WAPHL:
  - Nasal swab

- Throat swab
- Tracheal aspirate
- Bronchoalveolar lavage (BAL)
- Bronchial aspirate or wash
- Sputum
- Lung Tissue
- Viral culture

For more detail, see: Specimen Collection and Submission Instructions Influenza RT-PCR, Diagnostic

The following specimen types are preferred for H5N1(Avian Influenza) testing at WAPHL:

- If the patient does not have conjunctivitis (with or without respiratory symptoms),
  - Option 1:
    - Vial 1: Nasopharyngeal swab with
    - Vial 2: Nasal swab combined with an oropharyngeal swab (e.g., two swabs combined into one viral transport media vial).
  - Option 2: If these specimens cannot be collected, a single nasal, single nasopharyngeal, or single oropharyngeal swab is acceptable.
- If the patient does have conjunctivitis (with or without respiratory symptoms), one of the following pairs of specimens will be needed:
  - Option 1:
    - Vial 1: Conjunctival swab with
    - Vial 2: Nasopharyngeal swab
  - Option 2:
    - Vial 1: Conjunctival swab with
    - Vial 2: Nasal swab combined with an oropharyngeal swab
  - Option 3:
    - Vial 1: Conjunctival swab with
    - Vial 2: Nasopharyngeal swab with
    - Vial 3: Nasal swab combined with an oropharyngeal swab
- For novel or avian influenza serology, 5 cc separated serum (not whole blood). Testing to be performed at CDC, get approval of WA DOH Communicable Disease Epidemiology 206-418-5500.
- Patients with severe respiratory disease also should have lower respiratory tract specimens (e.g., an endotracheal aspirate or bronchoalveolar lavage fluid) collected, if possible. For severely ill persons, multiple respiratory tract specimens from different sites should be obtained to increase the potential for HPAI A(H5N1) virus detection.

For conjunctival swab collection instructions, see: Conjunctival Swab Specimen Collection for Detection of Avian Influenza A(H5) Viruses

For more information: Highly Pathogenic Avian Influenza A(H5N1) Virus: Interim Recommendations for Prevention, Monitoring, and Public Health Investigations | Bird Flu | CDC

Key points for specimen collection:

- Collect specimens using appropriate infection control procedures. **At a minimum** use droplet precautions. For suspected novel influenza use airborne precautions (face shield and N95 mask in addition).
- Collect nasopharyngeal, nasal, and throat specimens using swabs with a synthetic tip, such as Dacron or nylon, and a plastic or wire shaft. Specimens collected with cotton or calcium alginate swabs with wooden shafts will not be tested.
- Immediately after collection, place the swab or aspirate material into a sterile vial with 2–3 mL of viral transport media; for swab specimens, aseptically break or cut off the end of the swab shaft.

The shaft is most easily broken where it is scored.

- **Close vial tightly** to avoid leakage during transport.
- Do not let a swab come into contact with reagents used for other tests. If a swab contacts reagents for other tests, a new swab must be submitted.
- Label vial with patient's name AND a second identifier, specimen source, and date obtained.
- **Specimen Storage**: Optimal testing performance is obtained with freshly-collected specimens stored and shipped refrigerated (2–8°C) that arrive to the WAPHL for processing within 72 hours of collection. If you are unable to ship the specimen for testing within 72 hours of collection, any specimen except serum should be frozen at ≤ -70°C and shipped on dry ice.

When requesting testing through Lab Web Portal

- Please use Lab Web Portal (LWP) View File - MediaLab.
- For general subtyping requests per the HAN or an unsubtypeable result
  - Document in LWP as "unsubtypeable" for submissions from a tab that cannot subtype or a result of "unsubtypeable".
- Notification to the LHJ should be made only if the specimen is a suspected HPAI (H5) or Novel Influenza with epidemiologic risk factors or unsubtypable.
  - Document in LWP as "Suspect Novel Virus"

Storage, packaging, and shipping of specimens in viral transport media

All persons shipping packages containing medical specimens must have documented shipping training (USDOT and USPS Regulations for Packaging and Labeling Infectious Substances). For more information, phone the Virology Lab (206-418-5458) or see shipping guidelines here.

WAPHL is open to receive influenza specimens Mon – Fri 8am to 5pm. Special arrangements must be made with WAPHL for specimens to be received on weekends or holidays (please contact WAPHL at 206-418-5409). Specimens that arrive at WAPHL on Saturdays or holidays will be processed the next business day. If this will delay specimen for processing > 72 hours from collection, freeze specimen and ship on dry ice.

It is the responsibility as the shipper to correctly package and label specimens to meet shipping regulations.

Ship specimens to:

Washington State Public Health Laboratories
Attn: Virology Laboratory
1610 NE 150th Street Shoreline, WA
98155

When shipping influenza specimens please follow these steps:

- Check that the cap of the transport tube is securely closed; place tube in Biohazard Ziploc bag containing piece of super absorbent paper (bag and absorbent paper supplied with each Influenza Transport Kit).
- Ship according to PHL requirements. **Specimens will not be processed until ALL following information is known**:
  - Patient name, second identifier, and county of residence
  - Specimen type, date of collection and test requested
  - Submitter name, address, and telephone/FAX numbers
- Medical examiners see the Respiratory Panel Guidance for Medical Examiners
- Ensure patient's name and second identifier, are on specimen tube and match information on specimen submission form.

- Place up to five Biohazard Ziploc bags in the secondary container (e.g. 95 kPa bag or Tyvek bag, dependent on kit manufacturer).
- Place completed WAPHL Lab Web Portal Form in OUTSIDE of the secondary container. Forms are best kept in a Ziploc bag to protect from moisture.
- Place secondary container inside shipper with frozen ice packs if shipping cold. If shipping frozen, please use enough dry ice to keep specimens frozen overnight. Add sufficient packingmaterial (Styrofoam peanuts or other material) to prevent shifting of contents.
- Write shipper name/address on outside of the shipper. Be sure that the UN3373 label is fully visible.
- Choose shipping method for delivery ≤ 24 hours (e.g., FedEx, Greyhound, US Express Mail, private couriers). If using FedEx, shipper may use pre-paid FedEx air bill. Other shipping expenses paid by shipper. *FedEx Tip: Select FedEx Standard Overnight (will arrive by 10amnext day like FedEx Priority Overnight but is less expensive).*

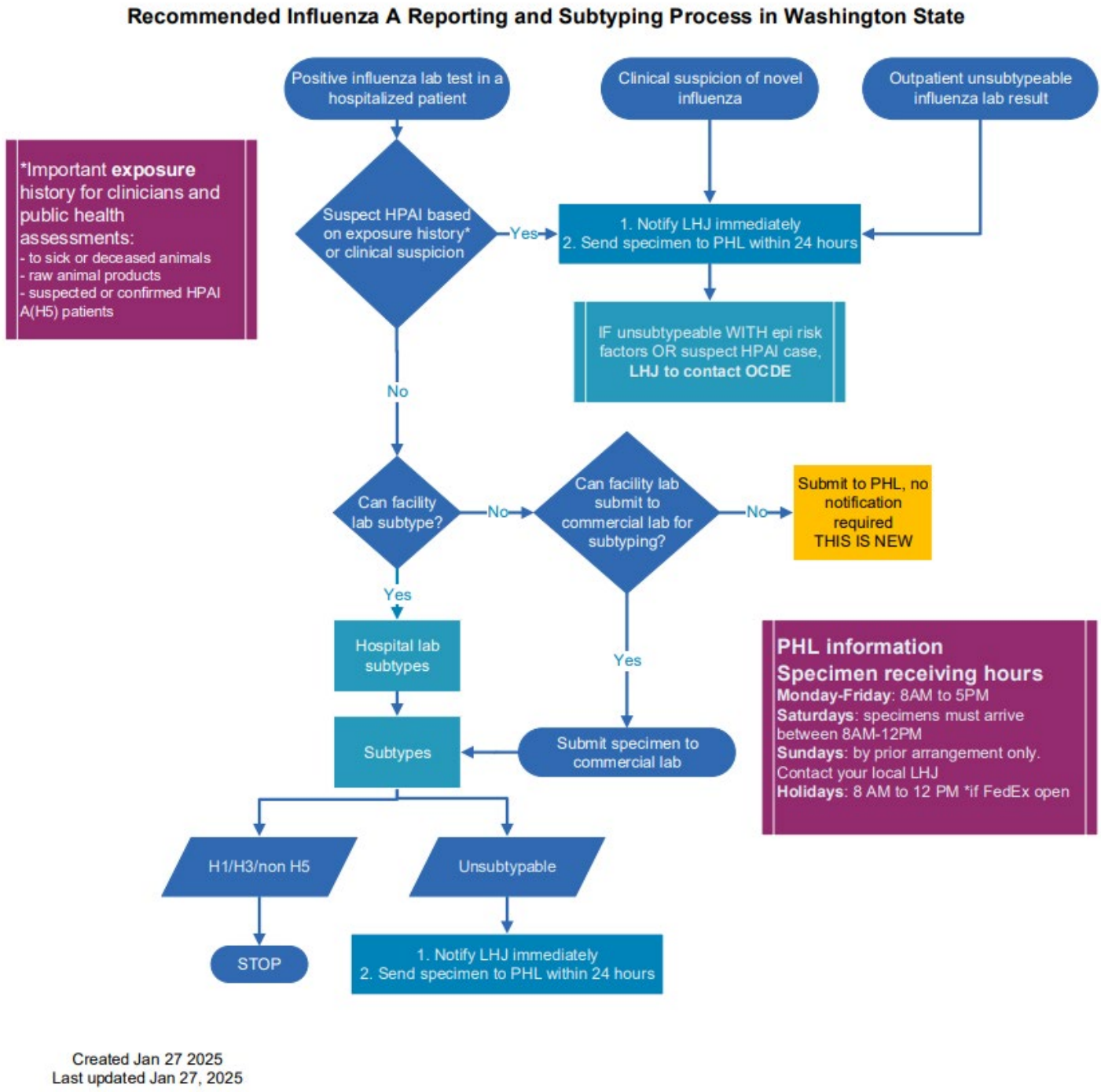
WAPHL testing procedures

Test results turnaround time: Projected turnaround time for influenza testing and subtyping using RT-PCR is up to 3 business days from specimen receipt.

Reporting of test results: Test results are sent by auto-fax to submitting facility. Test results will also be sent by fax and/or electronic reporting system to local health jurisdiction in which patient resides.

Subtyping Recommendations per CDC HAN, January 2025

For more information see: [Health Advisory: Subtyping of Influenza A Recommended for Hospitalized Patients | Washington State Department of Health]



QUESTIONS? Most questions should be directed to your local health jurisdiction.
Communicable Diseases Epidemiology may be reached at (206) 418-5500
WAPHL, Virology Laboratory may be reached at (206) 418-5458