



State of Louisiana
Department of Health and Hospitals
Office of Public Health Laboratories

Date: August 10, 2012

TO: Submitters of Influenza Specimens
Regional Medical Directors
Regional Administrators
Regional Nurse Managers

From: Danielle Haydel
Molecular/Virology Manager

RE: Influenza Surveillance and Influenza A(H3N2)v Testing

On August 3, 2013, CDC issued a health advisory notifying the public of an increase in Influenza A H3N2v virus infection in three U.S states. Contact with swine either directly or indirectly is still identified as the source of these infections. In the advisory, the following interim recommendations were made for health care providers.

- Clinicians who suspect influenza in persons with recent exposure to swine should obtain a nasopharyngeal swab or aspirate from the patient, place the swab or aspirate in viral transport medium, and contact their state or local health department to arrange transport and request a timely diagnosis at a state public health laboratory.
- Reverse-transcription polymerase chain reaction (RT-PCR) testing for influenza should be considered for patients with influenza-like illness prior to the start of the traditional influenza season in October.
- RT-PCR testing for influenza should be considered throughout the year for patients with influenza-like illness reporting recent swine exposure and for those who can be epidemiologically linked to confirmed cases of variant influenza.
- Commercially available rapid influenza diagnostic tests (RIDTs) may not detect H3N2v virus in respiratory specimens. Therefore, a negative rapid influenza diagnostic test result does not exclude infection with H3N2v or any influenza virus. In addition, a positive test result for influenza A cannot confirm H3N2v virus infection because these tests cannot distinguish between influenza A virus subtypes (they do not differentiate between human influenza A viruses and H3N2v virus). Therefore, respiratory specimens should be collected and sent for RT-PCR testing at a state public health laboratory.
- Clinicians should consider antiviral treatment with oral oseltamivir or inhaled zanamivir in patients with suspected or confirmed H3N2v virus infection. Antiviral treatment is most effective when started as soon as possible after influenza illness onset.

Since October 2011, the Louisiana Office of Public Health Laboratory has used the CDC Human Influenza Virus Real-time RT-PCR Diagnostic Panel to detect and characterize influenza from patient respiratory samples. With a new data interpretation update to this assay kit released by CDC on August 6, 2012, we can now provide a presumptive result of H3N2v at the state level before forwarding specimens on to CDC for further testing.

We ask that upon collection of samples for testing, you include relevant Epi Risk Factors on the lab submission form. Risk factors could include, but are not limited to, direct contact with swine, attendance at a fair or agriculture show or contact with a person previously diagnosed with H3N2v.

Specimen Collection and Transport

- **The requested specimen types are upper respiratory tract clinical specimens (including nasopharyngeal swabs [NPS], nasal swabs [NS], nasal aspirates [NA], nasal washes [NW] and dual nasopharyngeal/throat swabs [NPS/TS]) and lower respiratory tract specimens (including bronchoalveolar lavage [BAL], bronchial wash [BW], tracheal aspirate [TA], sputum, and lung tissue) from human patients with signs and symptoms of respiratory infection and/or from viral culture**
- Collecting the Specimen
 1. Follow specimen collection devices manufacturer instructions for proper collection methods.
 2. Swab specimens should be collected using only swabs with a synthetic tip, such as nylon or Dacron, and an aluminum or plastic shaft.
 3. Calcium alginate swabs are unacceptable and cotton swabs with wooden shaft are not recommended.
 4. Respiratory swabs should be collected and placed into viral transport media (VTM).
 5. For non-swab specimens, aseptically dilute liquid sample with an equal volume of viral transport media.
- **Store and ship specimens at 2-8°C. Wet ice transport is recommended.**
Transporting the Specimen
 1. Ensure that when transporting human respiratory specimens, all applicable regulations for the transport of the etiologic agents are met.
 2. Transport human respiratory specimens in VTM refrigerated at 2-8°C.
- **Samples must be received into the lab and processed within 72 hours of collection.**
Storing Specimens
 1. Store specimens refrigerated 2-8°C and ship to the State Laboratory on enough wet ice (a wet ice and freezer brick combination will work best during the warmer months) to maintain a 2-8°C transport temperature.
 2. Samples must be received and processed within 72 hours of collection. Ship specimens overnight to the State Laboratory on the day they are collected so we can process them prior to the 72 hour deadline.
 3. To minimize the effects of multiple freezing and thawing every attempt should be made to deliver the specimen to the laboratory within 72 hours from collection. If delivery to the laboratory within 72 hours from collection is not possible (ie. Sample collected on a Friday etc), freeze the specimen at $\leq -70^{\circ}\text{C}$ upon collection and ship to the laboratory on dry ice. If the sample is frozen at any point, it must remain and be shipped frozen. Document the date/time of freezing on the specimen submission form.

Laboratory Reporting

- **The laboratory will determine the best testing algorithm for your sample based on current reagent availability, epidemiologic data and specimen volume. Available options include:**

Influenza A/B Screen

1. InfA – universal detection of type A influenza viruses
2. InfB – universal detection of type B influenza viruses
3. RP – internal control measuring the effectiveness of our extraction procedure, the quality of the specimen and to allow us to detect PCR inhibition.
4. If influenza A is detected, the sample will automatically be retested for subtypes

Influenza A/B plus Subtyping

1. InfA – same as the InfA listed above
2. H1 – contemporary human A/H1 viruses
3. H3 – contemporary human A/H3 viruses
4. pdm InfA – nucleoprotein gene from 2009 H1N1 influenza virus
5. pdm H1 – hemagglutinin gene from 2009 H1N1 influenza virus
6. RP – same as the RP listed above

Specimens that have InfA, H3 and pdmInfA positive and H1 and pdmH1 negative are considered Presumptive Positive for Influenza A(H3N2)v Virus.

Influenza A/H5 Subtyping Assay

1. InfA, H5a, H5b and RP
 2. Testing for avian influenza A/H5N1 is considered on a case-by-case basis in consultation with the Infectious Disease Epidemiology department for hospitalized or ambulatory patients with:
 - a. Documented temperature of > 38°C AND
 - b. One or more of the following: cough, sore throat, shortness of breath, AND
 - c. History of contact with poultry or a known or suspected case of influenza A (A/H5N1) in an A/H5N1-affected country within 10 days of symptom onset.
- **Laboratory reports will have a list of the targets tested for on each specimen. Each influenza target that is tested will be resulted with Positive or Negative and there will be a final result interpretation printed at the bottom of the test section of the report.**

Report Exceptions

1. If the RP result is negative and no influenza targets are positive, upon a repeat test, the sample will be reported out as Inconclusive and you will be prompted to send another sample if additional testing is requested.
 2. RP results will not be listed on the final report.
- **Non-Standard result combinations such as being positive for Influenza A and Influenza B will have INCONCLUSIVE listed as the report conclusion. Additional comments may be added as warranted by the specific result combination.**

If you receive a report that you need additional explanation to understand, please contact either Kerri Gerage at 504-219-4646 or Danielle Haydel at 504-219-4670. If you have general questions about Influenza testing, please email Danielle Haydel at the email address listed below. Thank you for your attention.

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