



Steven H. Hinrichs, M.D., Director, Public Health Laboratory  
Thomas J. Safranek, M.D., Nebraska State Epidemiologist  
Samuel J. Pirruccello, M.D., Director, Client Services

981180 Nebraska Medical Center  
Omaha, NE 68198-1180  
Phone: (402) 559-2440  
Toll Free: 866-290-1406  
Fax: (402) 559-9497

Date: January 9, 2006

To: Nebraska Laboratories

From: Nebraska Public Health Laboratory

RE: Pandemic Influenza Laboratory Testing

Hello All:

The following questions and answers address the situation in which a specimen is received into your laboratory from an ill individual that has either documented travel to an area where avian influenza A H5 is/has occurred or has known exposure history (to an ill individual or bird) that would put them at high risk.

**1. How should doctor's offices, hospitals, or reference clinic laboratories handle specimens which get submitted to us to rule out avian flu?**

As per Centers for Disease Control and Prevention (CDC) guidelines, the laboratory should not perform any type of testing but should contact a NHHSS official to discuss this request. All NHHSS approved specimens will be forwarded to the NPHL for avian influenza A H5 testing on specimens *from initial or index cases* that are highly suspected of having avian influenza A H5.

**2. Will the NPHL perform the testing STAT?**

In the event of a possible H5 case, the rapid diagnosis of the disease agent will be critical. Therefore, this testing would be performed 24/7 at the NPHL upon request by authorized NHHSS officials.

**3. How much specimen will laboratories need to send to NPHL for testing?**

At a minimum, the NPHL should receive 2 – 3 ml of nasal washings. We should receive the entire specimen as laboratories other than the NPHL *should not do* any type of rapid tests (including DFA and EIA) or set up viral cultures if avian influenza A H5 is suspected.

**4. How long is going to take the NPHL to perform the testing?**

The CDC's testing protocol for avian influenza A H5 is performed on a PCR testing platform that utilizes the 96-well microtiter plate format. Typically one could expect results in as little as 8 to 12 hours once the specimen has been received into the lab. If repeat testing is needed, tests might take up to 24 hours to complete.

**5. Will the NPHL perform the DFA or an EIA rapid test for us so we will have a rapid (albeit preliminary) answer for our patient?**

As part of its testing panel, the NPHL will perform an EIA rapid influenza test even though it is realized that this test has sensitivity limitations. The influenza PCR testing panel will be performed upon initial receipt of the specimen. Additionally, the NPHL will set up viral cultures, including tissue culture tubes and spin-amplification shell-vials for the following respiratory viruses: Adenovirus, RSV, and Influenza A (generic) and B, and Para-influenza viruses 1, 2, 3. Results from the cultures could be known in as little as 24 hrs. Culture tubes will be held through 14 days. The NPHL will only set up cultures if the influenza PCR testing panel tests results are negative as we have been advised by the CDC not to perform cultures on specimens that are highly suspect to contain avian influenza A H5. Only the rapid non-culture tests- i.e. PCR, will be performed initially.



Steven H. Hinrichs, M.D., Director, Public Health Laboratory  
Thomas J. Safranek, M.D., Nebraska State Epidemiologist  
Samuel J. Pirruccello, M.D., Director, Client Services

981180 Nebraska Medical Center  
Omaha, NE 68198-1180  
Phone: (402) 559-2440  
Toll Free: 866-290-1406  
Fax: (402) 559-9497

**6. Would it be possible for those laboratories that perform viral cultures to go ahead and set these up and then wait for the answer from NPHL as to whether it is Avian flu or not?**

No. Taking into consideration the guidance provided by the CDC, laboratories other than NPHL should not to do any type of laboratory procedures with the specimen. The specimen should be sent to NPHL for preliminary testing. Again, NPHL should receive the entire specimen; laboratories other than NPHL should not set up viral cultures or run rapid tests.

**7. If the PCR test done at the NPHL is negative for H5, should the laboratory that submitted the original sample then set up a culture looking for the other respiratory viruses, or will NPHL do that?**

If the individual is highly suspect for having avian influenza A H5, the NPHL will run an influenza PCR panel using protocols provided by the CDC. The influenza PCR panel will include: Influenza A (generic), antigenic specific tests for Influenza A H1, H3, H5 and H7, as well as Influenza B. The NPHL would set up a viral culture if the influenza PCR panel test is negative. Cultures would be set up at the same time or right after the results of the panel were known. Culture results could be available as quickly as 24 hours. Other PCR-based testing may be indicated if the situation warranted.

**8. What testing could laboratories other than NPHL perform after cases of avian influenza H5N1 have been confirmed by the NPHL?**

If there are confirmed outbreaks (not individual cases or contained instances) occurring in Nebraska or elsewhere in the world, the WHO would issue updated guidance and could possibly place the global community on a Phase 4 (small clusters of ill individuals) or Phase 5 (larger clusters of ill individuals). A Phase 6 stage alert, signifying a true Pandemic Period, would be issued by the WHO if increased and sustained transmission in the general population was documented. If Phase 6 was achieved based on WHO criteria, the NHHSS and NPHL in conjunction with guidance from the CDC would recommend that hospital laboratories be allowed to perform EIA or DFA testing procedures using biosafety level 2 (BSL-2) containment protocols. BSL-2 protocols necessitate the use of Class II Type A biosafety cabinets and use of additional personal protective equipment including lab coats, gloves, and possibly N-95 respirator masks that laboratorians have been fit-tested and approved to use.

In summary, the NPHL has seen only "seasonal" or "normal" influenza activity to date; therefore, routine laboratory influenza testing can continue to be performed.

If the situation were to change and the reality that avian influenza A H5 were to become present in the US, the CDC would issue a Health Alert Network (HAN) to all state public health officials which would promptly be passed down to all health officials and laboratory staff throughout Nebraska.

If you have additional questions or comments, please contact Tony Sambol at [asambol@unmc.edu](mailto:asambol@unmc.edu) or 402-559-3032