



# **Guidelines for Sample Collection and Handling of Human Clinical samples for Laboratory Diagnosis of H1N1 Influenza**



**National Institute Of Communicable Diseases  
22- Sham Nath Marg New Delhi -110054**

***DIRECTORATE GENERAL OF HEALTH SERVICES  
MINISTRY OF HEALTH AND FAMILY WELFARE  
GOVT OF INDIA***

## **CASE DEFINITION OF H1N1 INFLUENZA**

A **suspected case** of swine influenza A (H1N1) virus infection is defined as a person with acute febrile respiratory illness (fever  $\geq 38^{\circ}\text{C}$ ) with onset.

- **within 7 days of close contact with a person who is a confirmed case of swine influenza A (H1N1) virus infection, or**
- **within 7 days of travel to areas where there are one or more confirmed swine influenza A(H1N1) cases, or**
- **resides in a community where there are one or more confirmed swine influenza cases.**

A **probable case** of swine influenza A (H1N1) virus infection is defined as a person with an acute febrile respiratory illness who:

- **is positive for influenza A, but unsubtypeable for H1 and H3 by influenza RT-PCR or reagents used to detect seasonal influenza virus infection, or**
- **is positive for influenza A by an influenza rapid test or an influenza immunofluorescence assay (IFA) plus meets criteria for a suspected case, or**
- **individual with a clinically compatible illness who died of an unexplained acute respiratory illness who is considered to be epidemiologically linked to a probable or confirmed case.**

A **confirmed case** of swine influenza A (H1N1) virus infection is defined as a person with an acute febrile respiratory illness with laboratory confirmed swine influenza A (H1N1) virus infection at WHO approved laboratories by one or more of the following tests:

- **Real Time PCR**
- **Viral culture**
- **Four-fold rise in swine influenza A (H1N1) virus specific neutralizing antibodies.**

**If any case fits in the case definition then samples should be collected according to the sample collection guidelines.**

### **What sample to be collected?**

Respiratory specimens including: bronchoalveolar lavage, tracheal aspirates, nasopharyngeal or oropharyngeal aspirates as washes, and nasopharyngeal or oropharyngeal swabs. Swab specimens should be collected only on swabs with a synthetic tip (such as polyester or Dacron) and aluminium or plastic shaft. Swabs with cotton and wooden shafts are not recommended. Specimens collected with swabs made of calcium alginate are acceptable.

### **When to Collect Respiratory Specimens?**

- As soon as possible after symptoms begin
- Before antiviral medications are administered
- Even if symptoms began more than one week ago
- Multiple specimens on multiple days could be collected if you have access to patient

**Specimen:** before initiating collection of sample a full complement of PPE should be worn.

### **Personal Protective Equipment**

- Masks (N-95)
- Gloves
- Protective eye wear (goggles)
- Hair covers
- Boot or shoe covers
- Protective clothing (gown or apron)

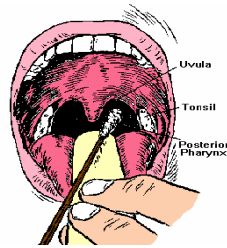
### **Methods of Collection**

- Throat swab
- Nasal / Nasopharyngeal swab

### **Throat Swab**

- Easy to do
- Highest yield in detecting H1N1 influenza in suspected cases
- Have the patient open his/her mouth wide open.

- The patient should try to resist gagging and closing the mouth while the swab touches the back of the throat near the tonsils.



**Nasal / Nasopharyngeal Swab:** Insert dry swab into nostril and back to nasopharynx. Leave in place for a few seconds. Slowly remove swab while slightly rotating it. Use a different swab for the other nostril. Put tip of swab into vial containing VTM, breaking applicator's stick.

**Nasal Swab** is collected from the anterior turbinate.



***Both Nasal and Throat swabs can be collected into the same VTM to increase the viral yield.***

### How to Label Samples

Use pre-printed barcode\* labels:

- On the specimen container
- On the field data collection form
- On the log book
- Subject's name
- Subject's unique identification number

<p><b><u>Label</u></b></p> <p><b><u>Specimen No. :</u></b></p> <p><b><u>Patient's Name :</u></b></p> <p><b><u>Hospital Name :</u></b></p> <p><b><u>Unique ID No. :</u></b></p>
--

### How to Store Specimens

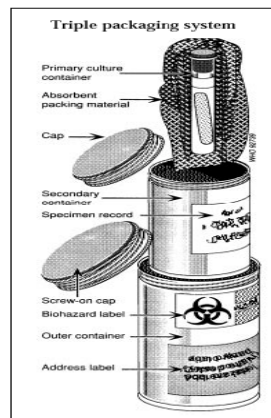
- Store specimens at 4 °C before and during transportation within 48 hours
- Store specimens at -70 °C beyond 48 hours

- Do not store in standard freezer – keep on ice or in refrigerator
- Avoid freeze-thaw cycles
- Better to keep on ice for a week than to have repeat freeze and thaw

## Transportation of specimens

Refer to WHO guidelines for the safe transport of infectious substances and diagnostic specimens

- Follow local regulations on the transportation of infectious material
- Coordinate with the laboratory



- All samples should be transported after proper packaging using the standard triple packaging system (WHO) and it should accompany with the clinical details as per proforma enclosed in **Annexure I**
- While transportation cold chain should be maintained

**Waste Disposal:** should be done as per guidelines of your hospital

Maintain adequately stocked specimen collection kits and store them properly when they are not in use.

Throat swabs are the easiest and best specimens to collect for suspected cases of avian influenza. Nasal swabs are easy to collect as well and should be done to increase yield.

Collect multiple specimens (respiratory and blood) on multiple days.

## General Biosafety Measures

- Clinical samples should be collected by hospital staff and not by the laboratory staff.
- All clinical samples have to be collected wearing complete complement of PPE.
- While taking samples always use N95 mask.
- Use Latex disposable gloves.
- Wear laboratory coat/disposable apron.
- Always cover your hairs with head cover.

- Use protective eye wear (goggles)/face shields
- The clinical samples should be processed only in designated laboratory having the appropriate containment facilities.
- All technical procedures should be performed in a way that minimizes the formation of aerosols and droplets.
- Adequate and conveniently located biohazard containers should be available for disposal of contaminated materials.
- Work surfaces must be decontaminated after any spill of potentially dangerous material and at the end of the working day. Generally, 5% bleach solutions are appropriate for dealing with biohazardous spillage. More information on disinfections and sterilization is provided in the WHO laboratory biosafety manual.
- Personnel must wash their hands often – especially after handling infectious materials and , before leaving the laboratory working areas, and before eating.
- Personal protective equipment must be removed before leaving the laboratory.

**NOTE: Whenever sample is send to laboratory(NICD, Delhi; NIV, Pune) a certificate should be attached with it stating that the sample is for research purpose and is packed properly and not hazardous to the community .**

