

Airborne Viruses

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Airborne Viruses

1. Influenza Viruses (Seasonal Influenza, Highly Pathogenic Avian Influenza - H5N1, Avian Influenza A – H7N9)
2. Novel Coronavirus (SARS/MERS)
3. Measles/Rubella Virus
4. Coxsackie Virus
5. Echoviruses
6. Parainfluenza Viruses
7. Respiratory syncytial Virus

Clinical Features

- Fever, chills, sorethroat, generalized muscular aches, malaise, headache, coughing, sneezing, running nose, pneumonia
- Rash (Measles/Rubella)

Laboratory Diagnosis

Clinical Specimen Sources

Be prepared to collect specimens before you leave for the field

- Suspected cases
 - Symptomatic cases
- Contacts
 - Including people living or working with suspected cases

The Specimen Collection Kit

Specimen Collection Kit

- Collection vials with VTM
- Polyester fiber-tipped applicators
- Tongue depressors
- Secondary container
- Ice packs (wet ice in sealed plastic bags)
- Cold box
- Zip-locked bags
- Personal protective equipment
- Field collection forms
- Permanent pen or marker for labeling samples
- Scissors

Personal Protective Equipment (PPE)

- Masks (N-95)



- Hair covers



- Gloves



- Boot or shoe covers



- Protective eye ware (goggles)



- Protective clothing (gown and apron)



How to Manage Kits

- Store specimen collection kits in a dry, cool place
- Store specimen collection kit where it will be accessible after office hours and on weekends

Specimen types

- Basically whether seasonal, avian, swine or pandemic, the samples for laboratory diagnosis are the same- respiratory specimens in **Viral Transport Media (VTM)**

Upper Respiratory tract specimens:

- Nasal swab
- Nasopharyngeal swab (**specimen of choice for MERS**)
- Nasopharyngeal aspirate
- Nasal wash
- Throat swab

Lower respiratory tract samples

- Transtracheal aspirate
- Bronchoalveolar lavage
- Lung biopsy
- Post-mortem lung or tracheal tissue

What to Collect

Preferred specimens

From an Ambulatory patient

- Nasopharyngeal swab *and*
- Throat swab
- Can be collected into the same VTM

From an Intubated patient

- Lower respiratory aspirate

Swabs – rayon, drayon, polyester swab



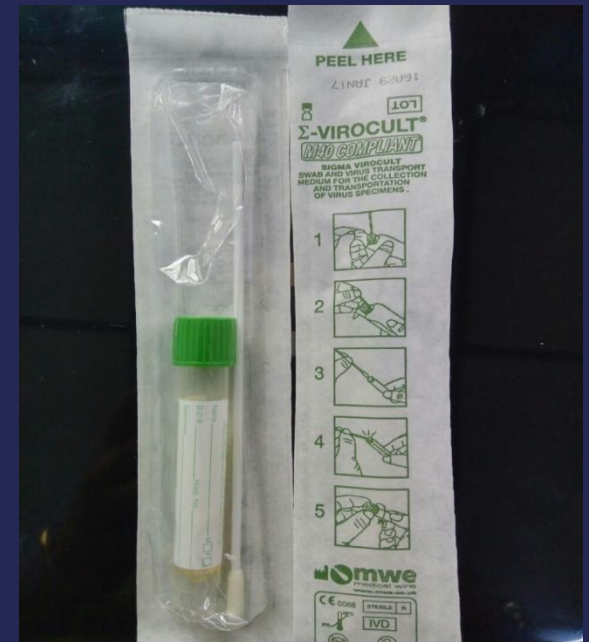
Cotton swab with wooden shaft – inhibit PCR

What is Viral Transport Medium?

- Used in the collection of samples for viral isolation and testing
- Prevents specimen from drying out
- Prevents bacteria and fungi growth

Storing VTM

- Sterile collection tubes containing 2-3 ml of VTM
- Tubes can be stored at 4-8 °C until use
- Tubes can be stored for short periods of time at Room Temperature
- Keep records of when the VTM tube was received and its expiry date



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
- Collect additional specimens when required or indicated

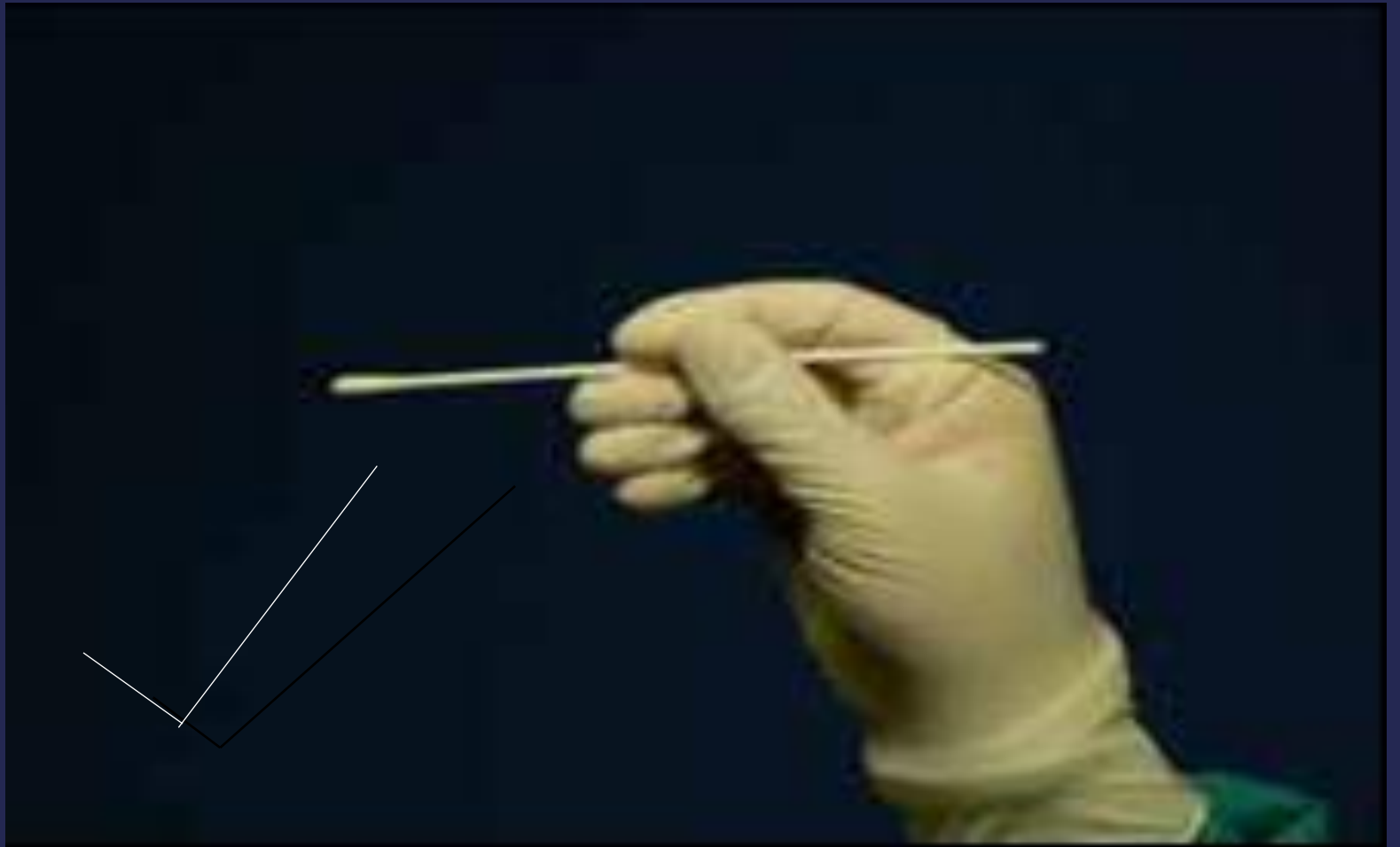
How to Collect Specimens

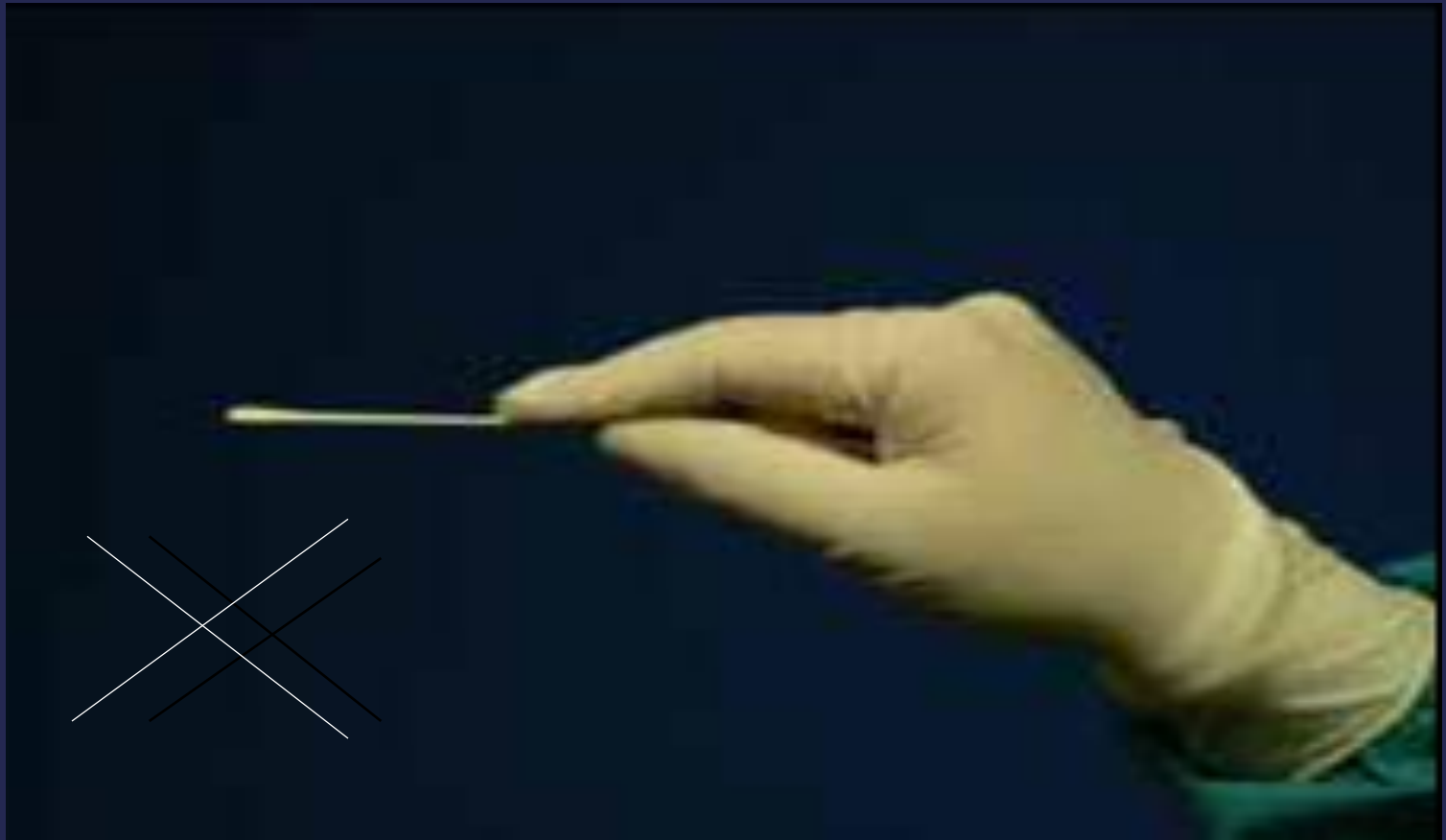
- Before collection - need to reassure and explain the procedure
- Children may need to be restrain
- Child's parents or guardian must be made aware that the child may become distressed
- Parents should not be in room- may interfere, more risk, no PPE



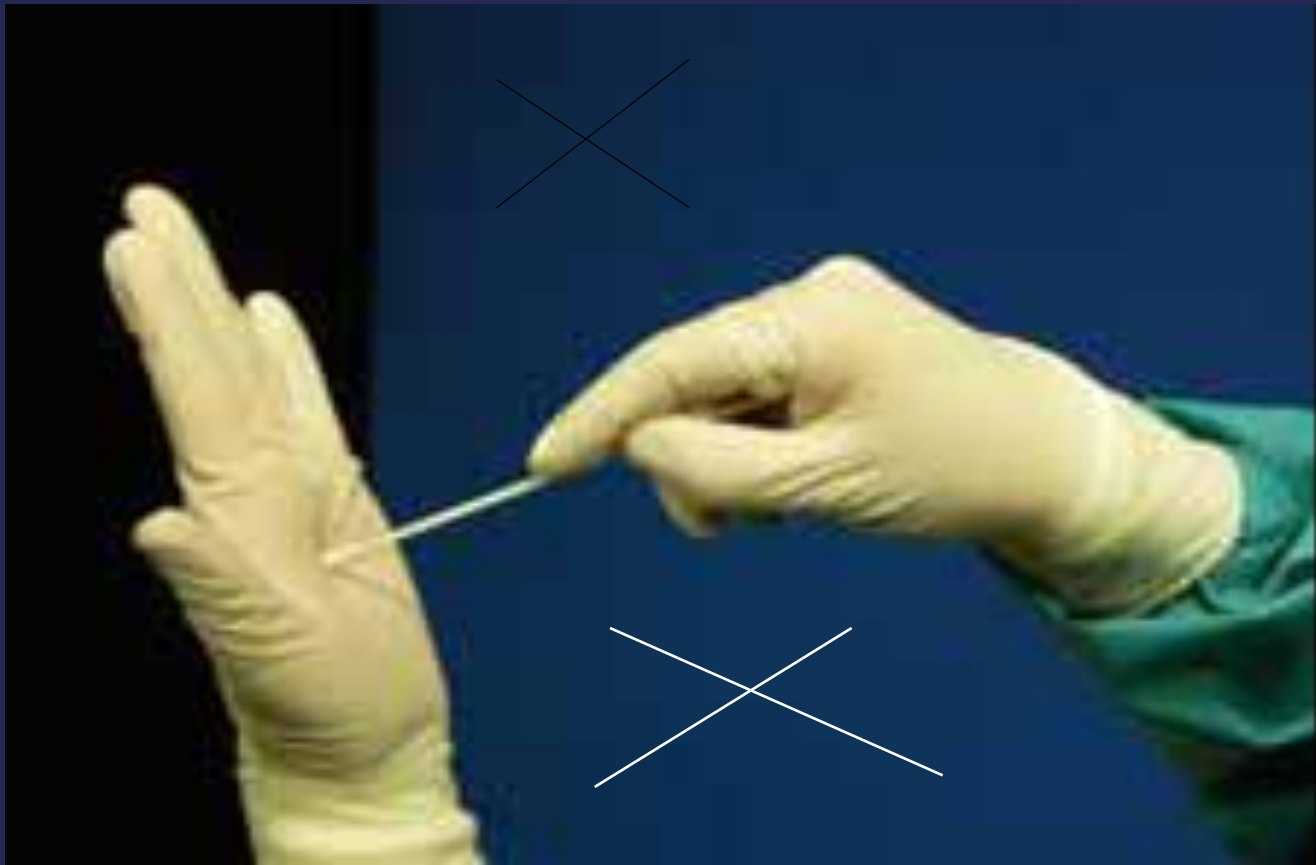






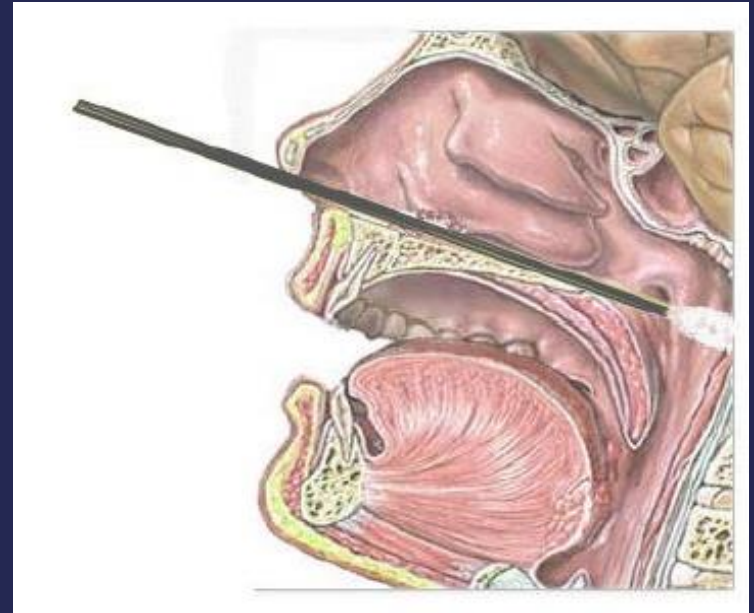






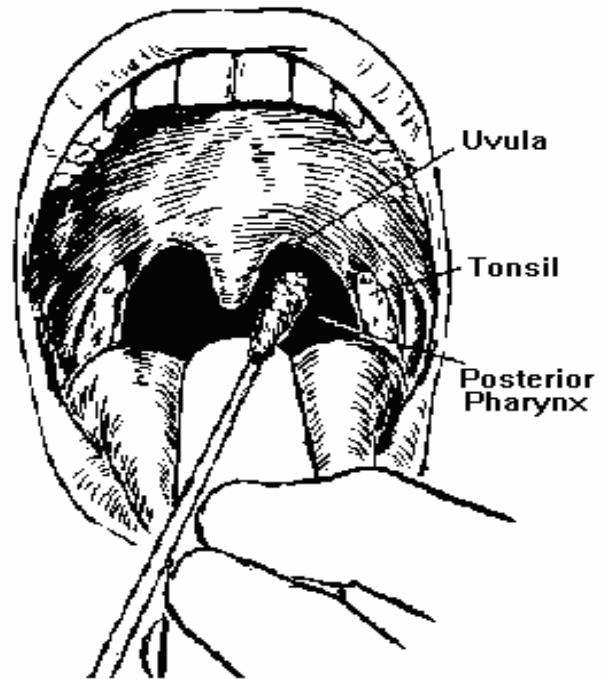
Nasopharyngeal Swab

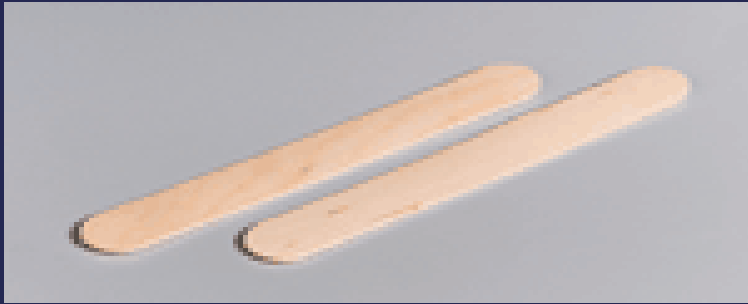
1. Insert dry swab into nostril and back to nasopharynx
2. Leave in place for a few seconds
3. Slowly remove swab while slightly rotating it
4. Put tip of swab into VTM tube, breaking applicator's stick
5. Use a different swab for the other nostril, same procedure as above



Throat Swab

- Tilt the patient's head back and gently depress the tongue with a tongue depressor
- The tonsillar areas and the posterior pharyngeal wall should be rubbed with the polyester swab to dislodge the epithelial cells
- Care should be taken not to touch the tongue and the lateral walls of the buccal cavity to avoid contamination with commensal bacteria
- After collection, break the shaft of the swab and place immediately into a VTM tube





Tongue Depressor



Throat Swab

Specimen Tracking System

Maintain a record to track:

- Identification number
- Subject information
- Specimen collection date
- Specimen collection location
- Diagnostic test results

Please complete this form carefully and circle the responses.

1. Report/Investigation Information: Name of Investigator(s): _____ Date - Case Reported: ____/____/____ Title / Office / Hospital : _____ Date - Case Investigated: ____/____/____ Notified by: _____																		
2. Case Identification: Patient's Name: _____ Date of Birth: ____/____/____ Age: years ____ months ____ Sex: ____ Father's Name: _____ Mother's Name: _____ Full Permanent Address: State/Region: _____ Township: _____ Village/ward: _____ Street No. & House No: _____ Phone No. _____																		
3. Hospitalization: Yes / No Date of Hospitalization: ____/____/____ Name of Hospital: _____ Hospital Registration Number: _____ Clinical Diagnosis: _____ (LI / SARI)																		
4. Immunization History: Vaccinated against Flu? Yes / No / Unknown Date of last Flu dose: ____/____/____																		
5. Signs and Symptoms: Date of onset of first symptoms : ____/____/____ Fever: Yes / No / Unknown Cough: Yes / No / Unknown Headache: Yes / No / Unknown Runny or Stuffy Nose: Yes / No / Unknown Muscle Ache: Yes / No / Unknown Sore throat : Yes / No / Unknown Joint Ache: Yes / No / Unknown Vomiting: Yes / No / Unknown Other symptoms: _____																		
Contact History : Yes / No / Unknown If yes, with whom: _____																		
MUI : Yes / No If yes, result of RDT : _____																		
6. Specimen Collection: <table border="1" style="width: 100%; border-collapse: collapse; margin-top: 5px;"> <thead> <tr> <th></th> <th>Date Collected</th> <th>Date Sent to Lab</th> <th>Date of Result</th> <th>Laboratory Results</th> </tr> </thead> <tbody> <tr> <td>Nasopharyngeal Swab</td> <td>____/____/____</td> <td>____/____/____</td> <td>____/____/____</td> <td>Positive / Negative</td> </tr> <tr> <td>Throat Swab</td> <td>____/____/____</td> <td>____/____/____</td> <td>____/____/____</td> <td>Positive / Negative</td> </tr> </tbody> </table>					Date Collected	Date Sent to Lab	Date of Result	Laboratory Results	Nasopharyngeal Swab	____/____/____	____/____/____	____/____/____	Positive / Negative	Throat Swab	____/____/____	____/____/____	____/____/____	Positive / Negative
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Nasopharyngeal Swab	____/____/____	____/____/____	____/____/____	Positive / Negative														
Throat Swab	____/____/____	____/____/____	____/____/____	Positive / Negative														
8. Case Classification: Lab confirmed Seasonal Influenza: / Lab confirmed Avian Influenza: / Discard																		
9. Signature of responsible person filling the form: _____																		

ILI Case Definition: An acute respiratory infection with: (1) measured fever of $\geq 38\text{ }^{\circ}\text{C}$; (2) and cough; (3) with onset within the last 10 days.

SARI Case Definition: An acute respiratory infection with: (1) history of fever or measured fever of $\geq 38\text{ }^{\circ}\text{C}$; (2) and cough; (3) with onset within the last 10 days; (4) and requires hospitalization.

Specimen Storage, Handling, and Transportation

How to Store Specimens

For specimens in VTM:

- Transport to laboratory as soon as possible
- Store specimens at 4 °C before and during transportation within 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

Specimen forwarding

Three main purpose

- To maintain specimen viability

- To prevent leakage outside the package

- To prevent cross contamination

*All samples for virus diagnosis must be sent in cold chain

Specimen transportation

Packing Specimens for Transportation

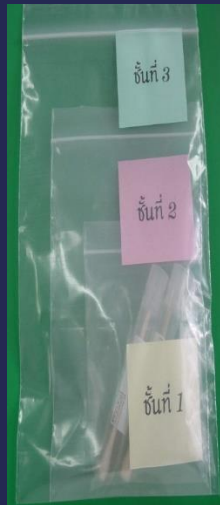
- Use three packaging layers
- First layer should be water tight
- Use absorbent material in all layers
- No more than 500 mL should be in the specimen container

Packing Specimens for Transportation

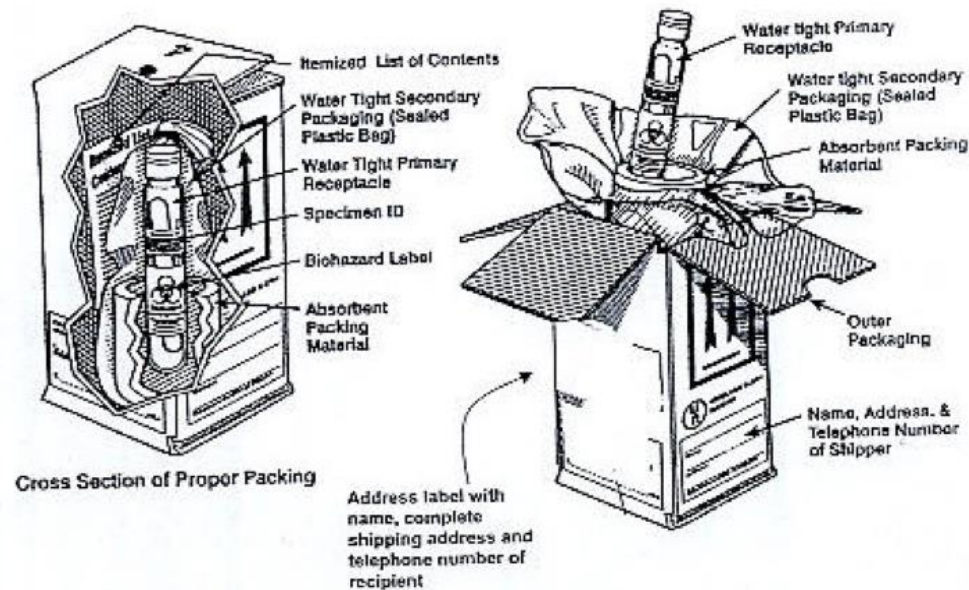
- Keep specimens at 4 °C
 - Fill a cooler with ice packs or coolant packs
- Include an itemized list of specimens with identification numbers and laboratory instructions

Transporting Specimens

- Refer to WHO guidelines for the safe transport of infectious substances and diagnostic specimens
- Inform and Coordinate with the laboratory
- NHL telephone: 01- 371957, 01- 371925



Transporting Specimens from Field to Lab

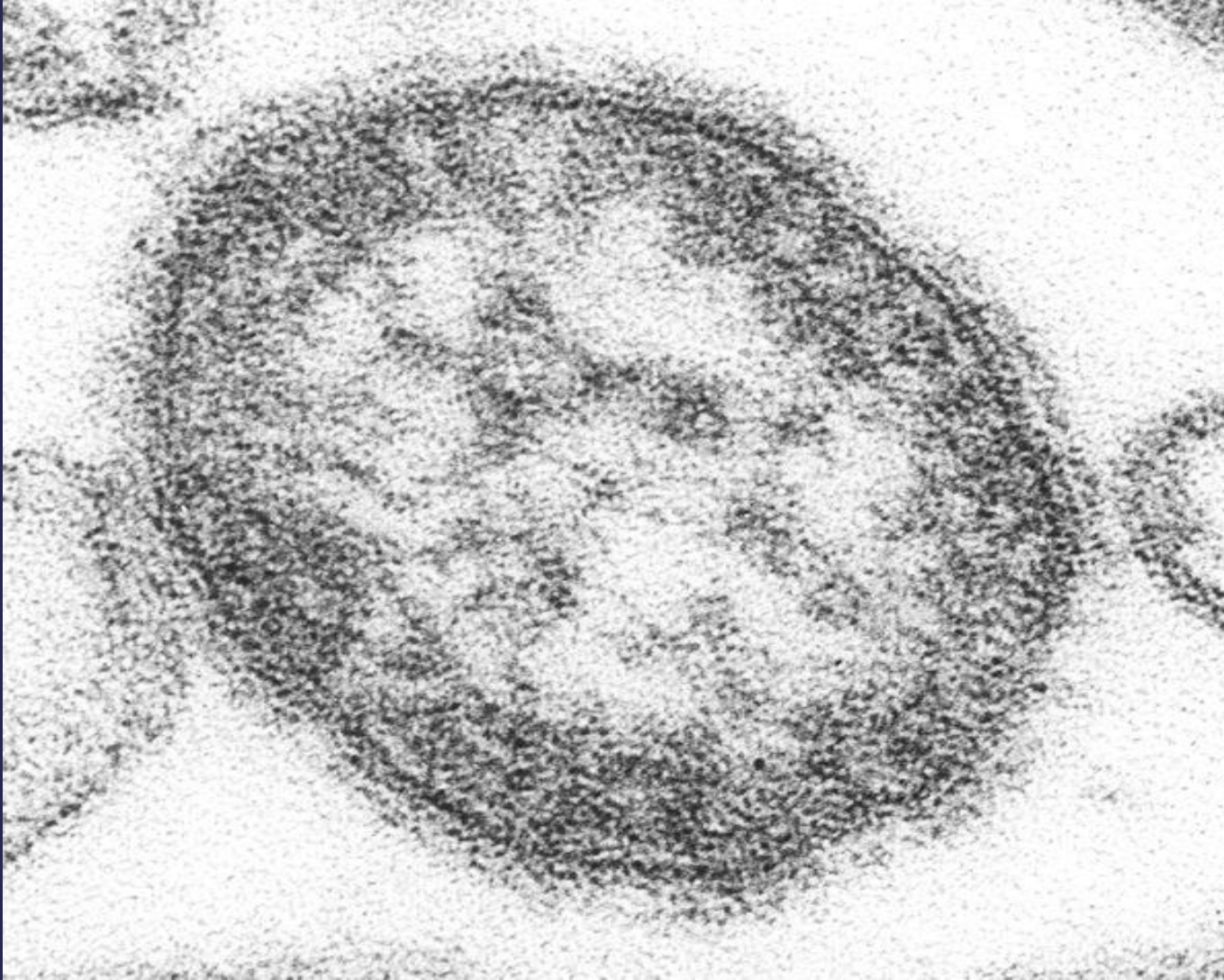


The labeling for contents should include the words:
“UN 3373 Diagnostic Specimens”



Measles Virus

Measles Virus



Transmission of infection

- Through the inhalation of infected aerosols and droplets
- Infected fomites are involved less frequently
- Highly communicable (99% chance of acquiring disease in non immune person)

Clinical Features

- Incubation period approx: 1 week to 10 days

- Clinical features are

fever ,

cough, coryza

conjunctivitis

} 3-4days

Koplik's spots (50-90%)

Rash

} 4—5 days

Koplik's spots

- Appear on the buccal mucosa
- Shortly before rash onset
- Small irregular red spots with a bluish white speck in the centre

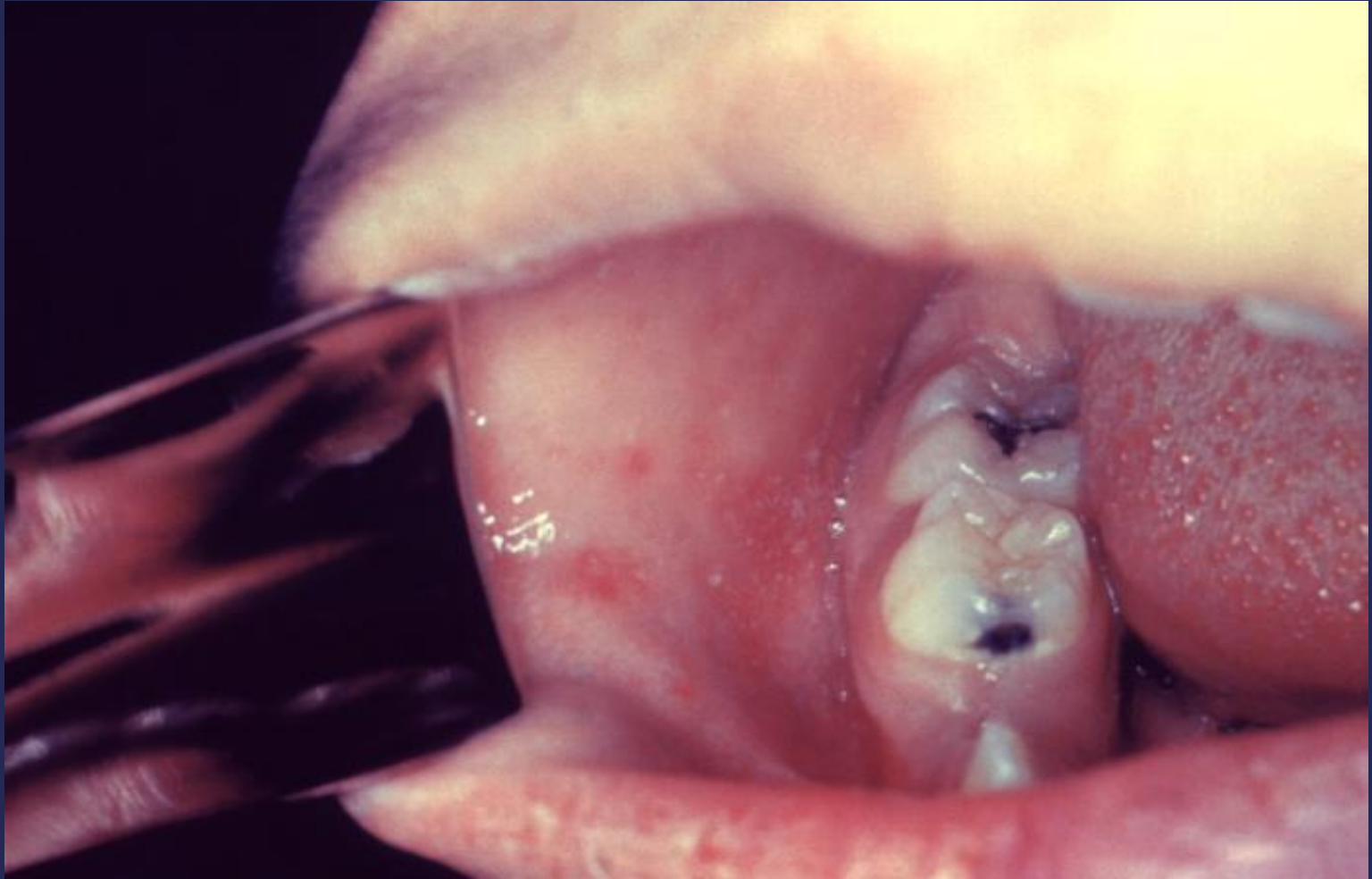
Koplik's Spots



Koplik's spots



Koplik's spots



Measles Rash

- First appear on the forehead or neck or behind the ears
- Lesions are red macules and become maculopapular
- By the end of second day upper extremities and trunk
- Third day lower extremities are affected

Measles



Measles Rash contd

- Rash resolves in the same order first disappearing from the face and neck
- last about 6 days
- turn brown and persists for 7-10 days
- Followed by fine desquamation

Complications

- Bronchitis, bronchiolitis, pneumonia and otitis media
- Encephalitis
- Diarrhoea
- Blindness
- Death- 1/1000 cases
- Risk of death is greater for infant and adult than children and adolescents

Laboratory Diagnosis

For Measles Virus culture,
Urine/ Throat swab are collected

1.Urine

- 10-20ml of urine collected in a sterile container
- First urine passed in the morning
- **Collect within 3 days after the onset of rash**
- Label the tube with the patient's name, outbreak ID number, specimen number, date of collection and specimen type

1.Urine

- Before transport, in the hospital laboratory, they should be kept at 4-8°C.
- Urine should be sent to NHL within 24 hours after collection (in cold box) with laboratory request form

2.Throat Swab

- **Collect within 3 days after the onset of rash**
- Tilt the patient's head back and gently depress the tongue with a tongue depressor.
- The tonsillar areas and the posterior pharyngeal wall should be rubbed with the polyester swab to dislodge the epithelial cells.
- Care should be taken not to touch the tongue and the lateral walls of the buccal cavity to avoid contamination with commensal bacteria

2.Throat Swab

- After collection, break the shaft of the swab and place immediately into a sterile leakproof container containing viral transport medium (VTM)
- Label the tube with the patient's name, outbreak ID number, specimen number, date of collection and specimen type.
- Before transport, in the hospital laboratory, they should be kept at 4-8°C.
- Throat swab should be sent to NHL within 48 hours after collection (in cold box) with laboratory request form.

For serology

- **Collect within 4 - 28 days after the onset of rash**
- Collect 5ml of blood in a sterile plain tube
- **one tube** is enough
- Test for Measles IgM Ab for recent infection
- Label the tube with the patient's name, age, sex, outbreak ID number, specimen number, date of collection and specimen type
- **We cannot do the samples without label on the tubes**

- Transport the whole blood specimen to NHL if it can reach within 24 hours.
- If it cannot reach NHL within 24 hours, do separation of serum
- Separate serum after clotting, and transfer into a new sterile bottle or microvial and send to NHL.
- To prevent insufficiency, collect **5 ml of blood or 2 ml of serum** in a sterile bottle
- For outbreak, **5 cases** enough
- Before transport, in the hospital laboratory, they should be kept at 4-8°C
- The specimens should be sent to NHL in cold box with laboratory request form

- The serum/ blood samples should not be haemolysed samples (Prevent hemolysis of samples – narrow needle, rapid suction, rapid pushing blood out of syringe, wet container should not be used)
- Measles Laboratory Requisition Form must include
 - Date of collection
 - Date of onset of rash
 - History of measles vaccination
 - Patient's address

Some of the lab forms are not filled completely. Please fill completely. Some samples are without lab request forms

Rubella Virus

Clinical and Virological Features

- Infects susceptible individuals via respiratory route
- Nasopharyngeal secretions- principle source
- Primary replication---epi cells of nasopharynx
- IP-14-21 days
- Viremia-widespread dissemination of the virus (blood, nasopharynx, urine, stool, synovial fl, skin, cervix, & L/N)
- Joint symptoms-commonest complication, appear soon after the rash faded, lasts for 3-4 days

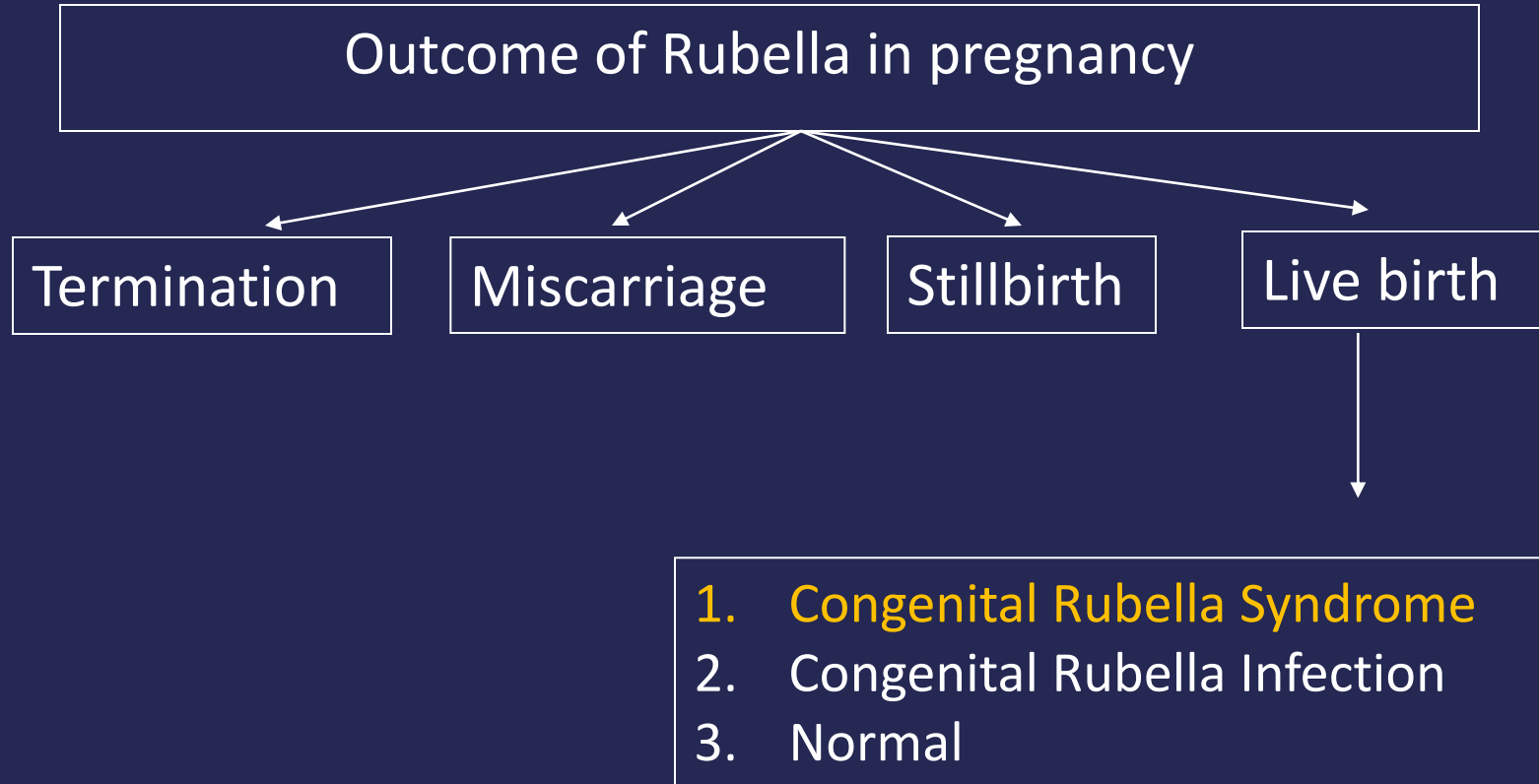
Clinical and Virological Features contd

- Patients- infectious for 3 wks----nasopharyngeal excretion may occur up to a week before the onset of rash & for 7-10 days thereafter
- Viremia is present about a week before the onset of rash, and end as rubella Ab develop
- 25%- inapparent infection
- Typical rubelliform rashes may result from infection with enteroviruses, human parvovirus B19 & some arboviruses (Chikungunya)

Rubella



Why is rubella infection so concerning ?



What is Congenital Rubella Syndrome (CRS)?

- A sequel of rubella infection in pregnancy
- Associated with Infection early in pregnancy
 - Weeks 1- 10 – 90% CRS*
 - Weeks 11-12– 33%
 - Weeks 13-14– 11%
 - Weeks 15-16– 24%
 - Weeks \geq 17– 0%

* Miller E, Cradock-Watson JE, Pollock TM. Consequences of confirmed maternal rubella at successive stages of pregnancy. Lancet. 1982 Oct 9;2(8302):781-4. PubMed PMID:6126663.

The purpose of rubella vaccination program is thus prevention of congenital rubella infections which can lead to fetal deaths/loss, pre-mature delivery or CRS

Congenitally Acquired Infection

- Rubella in pregnancy--fetal death and spontaneous abortion or delivery of a severely malformed infant, an infant with minimal damage or a healthy infant
- Outcomes depend on gestational age at which maternal infection occurs
- First 8 weeks of pregnancy- spontaneous abortion in 20% of cases
- 13th-16th weeks of gestation- 17% of infant may develop deafness & retinopathy

Congenitally Acquired Infection contd

- Rubella virus can be recovered from most infants with severe congenitally acquired rubella at birth
- 3 months of age----50-60% of nasopharyngeal secretions
- 9-12 months of age---10%
- Delayed manifestation- diabetes mellitus & other endocrinopathies, sensory neural deafness, glaucoma and progressive panencephalitis

How does CRS present clinically?

Organ specificity generally related to stage of gestational infection.

PERMANENT

- Hearing Impairment
- Ophthalmologic
 - Cataract, Microphthalmia, Retinopathy, Glaucoma
- Heart defects
 - Patent Ductus Arteriosus
- Microcephaly
- Developmental Delay

TRANSIENT

- Thrombocytopenia
- Jaundice
- "Blueberry muffin" appearance
- Hepatosplenomegaly
- Bone lucencies

DELAYED

- Endocrinopathies
- Progressive auditory or ocular dysfunction



Thank you for your kind attention

<http://go.funpic.hu>