Orthomyxoviridae
Paramyxoviridae

Assistant Professor Dr. Mushtak Talib S. Al-Ouqaili
Orthomyxovirus (Influenza) Family

The name myxovirus was originally applied to influenza viruses. It meant virus with an affinity for mucins. Now, there are 2 main groups – the orthomyxoviruses and the paramyxoviruses.

Differences between orthomyxoviruses and paramyxoviruses

<table>
<thead>
<tr>
<th>Feature</th>
<th>Orthomyxoviruses</th>
<th>Paramyxoviruses</th>
</tr>
</thead>
<tbody>
<tr>
<td>Viruses and diseases</td>
<td>Influenza A,B,C</td>
<td>Mumps, measles, respiratory syncytial, para-influenza</td>
</tr>
<tr>
<td>Genome</td>
<td>Single-stranded RNA in 8 pieces, MW 2-4x10^6</td>
<td>Single-stranded RNA in single piece, MW 5-8x10^6</td>
</tr>
</tbody>
</table>
ORTHOMYXOVIRUSES (INFLUENZA VIRUSES)

Classification

- **Type A** viruses cause the most cases of influenza in humans and undergo mutations more frequently than the other type viruses…

- **Type B** viruses are endemic in USA and associated with local epidemics…

- **Type C** viruses rarely cause disease
Orthomyxoviruses. Nomenclature

Influenza type  →  Year of isolation  →  Hemagglutinin subtype

Geographic source  ↓  Isolate number  ↓  Neuraminidase subtype

A/Sydney/5/97 (H3N2)
Orthomyxoviruses: medium-sized, enveloped, (-) sense that vary in shape from spherical to helical. Their genome is segmented into eight pieces.
Influenza viral genome
✓ (-) ssRNA
✓ 8 segments (pieces)
✓ One gene per segment
✓ nucleoprotein
✓ matrix proteins
✓ subunits of RNA polymerase
✓ spikes (about 500)

Flu viruses are named by the type of surface proteins

- Hemagglutinin - trimer (HA)
  ✓ Helps virus enter cell
  ✓ Type A infects humans, birds and pigs
- Neuraminidase - tetramer (NA)
  ✓ Helps virus exit cell
  ✓ 9 subtypes

Flu Viruses Currently infecting...
✓ Humans: H1N1, H1N2, and H3N2
✓ Avian Flu Virus: H5N1
ORTHOMYXOVIRUSES

HA - hemagglutinin
NA - neuraminidase
helical nucleocapsid (RNA plus NP protein)
lipid bilayer membrane
polymerase complex
M1 protein

type A, B, C: NP, M1 protein
sub-types: HA or NA protein
Haemagglutinin (HA)
Encoded by RNA segment # 4
Can agglutinate red blood cells - hence the nomenclature
Cleavage by host-cell protease is required (resulting in HA<sub>1</sub> and HA<sub>2</sub>) for infection to occur
Hemagglutinin glycoprotein is the viral attachment protein and fusion protein, and it elicits neutralizing, protective antibody responses

Neuraminadase (NA)
Encoded by RNA segment # 6
Removes neuraminic (sialic) acid from cell and permits dissemination of viruses
Important in releasing mature virus from cells
Stimulates production of protective antibodies
Influenzavirus B

- Virions enveloped
- About 500 spikes
- Nucleocapsid enclosed within lipoprotein membrane
- Virions contain 8 segments of linear negative-sense single stranded RNA
- Total genome length is 13588 nt
- The largest segment 2341 nt

- Infect much man and birds.
- Cause human disease but generally not as severe as A types.
- Believed to be epidemiologically important - reassortment with type A leads to epidemics.
Antigen

Influenza viruses are divided into 3 groups determined by the ribonucleoprotein (RNP) antigen and M antigen.

1-Soluble antigens: include which are much stribonucleoprotein and M protein variable in antigenicity.

2-Surface antigens: include HA and NA which are much variable in antigenicity.
## Features of viral genera

<table>
<thead>
<tr>
<th>Feature</th>
<th>TYPE A</th>
<th>TYPE B</th>
<th>TYPE C</th>
</tr>
</thead>
<tbody>
<tr>
<td>severity of illness</td>
<td>++++</td>
<td>++</td>
<td>+</td>
</tr>
<tr>
<td>animal reservoir</td>
<td>yes</td>
<td>no</td>
<td>no</td>
</tr>
<tr>
<td>human pandemics</td>
<td>yes</td>
<td>no</td>
<td>no</td>
</tr>
<tr>
<td>human epidemics</td>
<td>yes</td>
<td>yes</td>
<td>no (sporadic)</td>
</tr>
<tr>
<td>antigenic changes</td>
<td>shift, drift</td>
<td>drift</td>
<td>drift</td>
</tr>
<tr>
<td>segmented genome</td>
<td>yes</td>
<td>yes</td>
<td>yes</td>
</tr>
<tr>
<td>amantadine, rimantidine</td>
<td>sensitive</td>
<td>no effect</td>
<td>no effect</td>
</tr>
<tr>
<td>zanamivir</td>
<td>sensitive</td>
<td>sensitive</td>
<td></td>
</tr>
<tr>
<td>surface glycoproteins</td>
<td>2</td>
<td>2</td>
<td>(1)</td>
</tr>
</tbody>
</table>
1. Attachment to the epithelial cells of the host through hemagglutinin.
2. Endocytosis
3. Uncoating
4. The RNA enters the nucleus of the cell where fresh copies are made.
5. These copies return to the cytosol where some serve as mRNA molecules to be translated into the proteins of fresh virus particles.
6. Progeny virions are formed and released by budding from the plasma membrane of the cell (aided by the neuraminidase) thus spreading the infection to new cells.
Need to make mRNA

MINUS (NEGATIVE) SENSE RNA GENOMES

proteins

(+ve) sense mRNA

(−ve) sense genomic RNA
Need to make mRNA

MINUS (NEGATIVE) SENSE RNA GENOMES

RNA polymerase must be packaged in virion.

If used, RNA modifying enzymes are packaged in virion.
1918 Influenza epidemic

- 20 million died of the flu
- A new influenza vaccine must be developed yearly
Mechanisms of Influenza Virus Antigenic “Shift”

15 HAs
9 NAs

Non-human virus

DIRECT

Human virus

Reassortant virus
Antigenic changes of Influenza A

- Viruses can undergo frequent changes due to recombination, reassortment, insertions and point mutations
  - Antigenic drift
  - Antigenic shift occurs every 8-10 yrs
  - Minor antigenic changes favor persistence of the viruses in the population and allow recombination that can eventually lead to severe epidemics and/or pandemics
ANTIGENIC DRIFT

✓ Gradual accumulation of mutations that allow the hemagglutinin to escape neutralizing antibodies
✓ Epidemic strains thought to have changes in three or more antigenic sites

GRADUAL ANTIGENIC CHANGE WITHOUT A CHANGE IN SUBTYPE

H3N2 → H3N2 → H3N2 → H3N2
1968 → 1975 → 1993 → 2004
HONG KONG → VICTORIA → BEIJING → FUJIAN
Antigenic drift

- Antigenic differences can result from changes in one amino acid
- Can involve any antigenic protein
- Can occur every year

- RNA replication is error prone
- New HA types are created frequently
- Requires new vaccine every “season”
Antigenic shift

- Occurs every 8-10 yrs
- Major antigenic change of either H or N antigens or both H and N
- Occurs by gene reassortment after simultaneous infection of a cell with two different viruses
- Three different H proteins and 2 major N proteins have evolved

<table>
<thead>
<tr>
<th>HөNө Spanish flu</th>
<th>HөNө Avian flu</th>
<th>HөNө Asian flu</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>HA</td>
<td>HA</td>
<td></td>
</tr>
<tr>
<td></td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td></td>
<td>HA</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
What is an Epidemic?

- The occurrence of more cases of disease than expected in a given area or among a specific group of people over a particular period of time*.

What is a Pandemic?

An epidemic occurring over a very wide area (several countries or continents) and usually affecting a large proportion of the population.

Examples:
- Cholera
- AIDS
- Pandemic Influenza
Where does influenza come from?

Type A constantly circulates in natural reservoirs

- Birds are the natural reservoir of all subtypes of Influenza A viruses
  - Migratory waterfowl
  - Chickens, turkeys, ducks, geese
- Humans
- Pigs
- Horses
- Other
Why do we not have influenza B pandemics?

- so far no shifts have been recorded
- no animal reservoir known
Epidemiology

- Source of infection: patients and carriers.
- AEROSOL
  - 100,000 TO 1,000,000 VIRIONS PER DROPLET
- Common: large droplets (sneezing, coughing, contact with saliva)
- Probably common: contact
  - Direct
  - Fomite
- Rare: airborne over long distance
- 18-72 HR INCUBATION
SYMPTOMS

- FEVER
- HEADACHE
- MYALGIA
- COUGH
- RHINITIS
- OCULAR SYMPTOMS
- CHILLS and/or SWEATS

Infection may be very mild, even asymptomatic, moderate or very severe
Clinical Responses

- Acute Symptoms last one week
  - Abrupt onset of fever, myalgia, headache and non-productive cough
- Fatigue and weakness can last 2-3 weeks.
- Infected individual predisposed to bacterial infections – *Staphylococcus*, *Streptococcus*, *Hemophilus*
- Other complications - **Reyes Syndrome**
- Immunity dependent upon localized anti-viral secretory IgA (strain specific)
- Develop long lasting circulating anti-viral IgG
Immunity to influenza

- Antibody to HA - > protective
- Antibody to NA - > decrease severity
- Serum antibody - > years
- Secretory antibody - > months
Laboratory Diagnosis

VIROLOGICAL

Respiratory secretions (direct aspirate, gargle, nasal washings)

- Virus isolation and growth in embryonated eggs
- Cell culture in primary monkey kidney or madindarby canine kidney cells
- Hemagglutination (inhibition)
- Hemadsorption (inhibition)
- IFA/ ELISA
- Direct immunofluorescence
Laboratory Diagnosis

Serodiagnosis

*Four-fold or greater increase in hemagglutination inhibition antibody titers between acute and convalescent specimens*

- Hemagglutination inhibition
- Hemadsorption inhibition
- ELISA
- Complement fixation test
- NT
Prophylaxis
Masks and Hand Washing

- Hand washing
  - Generally perceived to be useful
  - No studies specifically performed for influenza
  - Easy to recommend

- Masks
  - Effectiveness not shown for influenza
  - However, could reduce transmission associated with large droplets

To be Continued…
Types of Vaccine

- **Killed Whole Virus**
inactivated virus vaccine grown in embryonated eggs; 70-90% effective in healthy persons <65 years of age, 30-70% in persons ≥65 years

- **Live Virus**
Attenuated strains were widely used in Russia but not elsewhere.

- **Virus Subunit**
HA extracted from recombinant virus forms the basis of today's vaccines.

- **Synthetic**
Much research is being done to try and find a neutralising epitope that is more stable, and can therefore be used for a universal vaccine.
Prevention and Treatment

70-90% effective in preventing illness

- **RIMANTADINE** (blocks the M2 ion channel) \((M2)\)
  - type A only, needs to be given early
- **AMANTADINE** (blocks the M2 ion channel) \((M2)\)
  - type A only, needs to be given early
- **ZANAMIVIR** (neuraminidase inhibitors) \((NA)\)
  - types A and B, needs to be given early
- **SELTAMIVIR** (neuraminidase inhibitors) \((NA)\)
  - types A and B, needs to be given early