Influenza Viruses – A Review

AVIAN INFLUENZA: INTERSECTORAL COLLABORATION
Larnaca, Cyprus
20 – 22 July 2009
Influenza Viruses

C. Goldsmith, 1981
Influenza Viruses

- Family *Orthomyxoviridae*, three influenza genera
  - Type A
  - Type B
  - Type C

C. Goldsmith and A. Balish, 2009
Influenza B

- Not classified into subtypes
- Mostly causes disease in humans
- It is a common influenza virus
  - Included in human seasonal influenza vaccine
- In general
  - Less severe than A
  - Epidemics occur less often than A

C. Goldsmith, 2005
Influenza C

- Not classified into subtypes
- Identified in humans and swine
- Different pattern of surface proteins
- Rare infections, with mild to no symptoms

C. Goldsmith, 2005
Influenza A Viruses

C. Goldsmith, 1981
Influenza A

- Negative single-stranded RNA virus
- 8 gene segments code for 11 proteins
- Most virulent
- Widest host range

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Influenza A

HA – receptor binding and fusion to host cell to support infection

NA – supports release of new virus from infected cell

http://en.wikipedia.org/wiki/Influenza_virus
Influenza A

Matrix
M1 – assists in viral assembly
M2 – provides low pH during viral synthesis

http://en.wikipedia.org/wiki/Influenza_virus
Influenza A

PB1, PB2, PA – polymerase complex for transcription

http://en.wikipedia.org/wiki/Influenza_virus
Influenza A

NP – nucleoprotein, supports viral synthesis

http://en.wikipedia.org/wiki/Influenza_virus
Influenza A

NS1, NS2 – Nonstructural protein, multifunction, downplays host immune response, assists in viral assembly

http://en.wikipedia.org/wiki/Influenza_virus
Influenza A

- Influenza A viruses
  - Categorized by subtype of HA and NA
  - Hemagglutinin (HA)
    - H1 – H16
    - Antibody to HA is protective
  - Neuraminidase (NA)
    - N1 – N19
    - Antibody to NA can help modify disease severity

Hemagglutinin
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Influenza A Virus Nomenclature

Virus type: A /Duck/ Sydney / 05 / 97 (H3N2)

Type of Animal (Optional): Duck

Place virus isolated: Sydney

Year isolated: 05 / 97

Virus subtype: H3N2
Laboratory Testing for Influenza A Humans

• Rapid diagnostic tests
  • Can provide results <30 minutes
  • ~ 70+% sensitive, 90+% specific

• Serology
  • Must used paired serum samples
  • >2 week delay for results

• Viral culture
  • Gold standard
  • Results take 7 days
  • Influenza isolates for yearly vaccine development

• RT-PCR
  • Most sensitive
  • Becoming more widely available

• Immunofluorescence
  • Requires intact cells and laboratory skill/experience
Laboratory Testing for Influenza A Animals

  - Chapter 2.3.4. Avian influenza
  - Chapter 2.5.7. Equine influenza
  - Chapter 2.8.8. Swine influenza

- OIE/FAO Network of Expertise on Animal Influenza (www.offlu.net)
Influenza A Epidemiology

Tumpey and Goldsmith, 2009
Influenza A Viruses

- Polymerase complex lacks proofreading capability → 1 in 5 virus particles likely to contain a mutation.
- If mutation provides virus with a competitive advantage, that strain quickly replaces its predecessor.
- In humans, the need to escape preexisting immunity exerts positive selection pressure on changes in amino acids comprising the antigenic sites of HA and NA.
- This antigenic drift results in the emergence of an antigenically distinct variant strain in humans every 2–3 years.
Influenza A Viruses

• Mutate frequently – escaping immunity
  • Antigenic drift
  • Point mutations accumulated during virus replication
  • Antigenic shift
    • Hybrid virus emerges when a cell is infected with two different influenza viruses
      – Human, avian, swine, equine
  • Transfer of influenza virus to a different species
Zoonotic Pathways of Influenza A Viruses

History of Animal (and Zoonotic) Influenza A Viruses

- 1930 - Swine Influenza Virus isolated (A/swine/Iowa/30)
- 1933 – Human Influenza Virus isolated
- 1902/1955 Avian (chicken) influenza virus isolated (A/chicken/Brescia/1902 [H7N7])
Influenza of Wild Birds

- Influenza virus replicates in respiratory and particularly gastrointestinal tracts
- Large amount of virus can be shed in feces
- Efficient viral transmission through fecal-oral route through contaminated water
- Certain species (waterfowl) believed to be natural reservoir
- All the HA and NA subtypes have been identified among wild birds – historically as asymptomatic infections
- Several species capable of distributing virus across countries / continents
- Influenza A, HPAI H5N1 is different
Influenza of Domestic Birds

- Domestic poultry (e.g., chicken, turkeys, ducks, etc.) and caged pet birds
  - Viruses can be divided into two groups based on their clinical manifestation in chickens
    - Highly pathogenic (within the H5 and H7 subtypes, to date) → severe systemic illness, up to 100% mortality
    - Low pathogenic (can also include H5 and H7 subtypes) → milder, primarily respiratory disease; may mutate into HP virus
  - Both likely introduced into domestic poultry from wild birds
Influenza of Domestic Birds

• Spread within and among flocks by various methods
  • Migratory birds
  • Mechanical transfer of infective feces
  • Contaminated water
  • Illegal trade
Influenza A of Swine

• Respiratory illness described in pigs in 1918 – believed to be the first occurrence of influenza in swine
• “Classical” swine influenza
  • Swine influenza virus first isolated as H1N1
  • Swine influenza endemic in many pig populations
  • Pigs can be infected with different influenza viruses - “reassortant strains”
Receptor Binding

• Influenza viruses bind to neuraminic acids (sialic acids) on the surface of cells to initiate infection and replication.

• HAs of viruses that replicate in different species show specificity toward sialic acids with different linkages:
  • Human viruses preferentially bind to receptor by an $\alpha 2,6$ linkage (SA$\alpha 2,6$Gal) - human tracheal epithelial cells contain mostly SA$\alpha 2,6$Gal.
  • Avian viruses mostly bind to sialic acid with an $\alpha 2,3$ linkage - gut epithelium from ducks possesses mostly SA$\alpha 2,3$Gal.
  • $\alpha 2,3$ linkages are present in the human airway epithelium cells that can be infected with avian influenza viruses.
  • Ability to infect is not solely based on these receptor types.

• Viruses passed through a particular host, they can adapt to that host by mutating the HA receptor-binding site.
Receptor Binding – and the Potential Role of Pigs as Mixing Vessels

• Swine have both SAα2,3Gal and SAα2,6Gal
• They can be infected not only with swine influenza viruses, but also avian and human
• Cells with receptors types found in different regions of the respiratory tract
• Same cell infected with human, avian, and/or swine viruses can support reassortment
Reassortments among Influenza A Viruses Infecting Swine

### Enzootic Strains among Swine, 2000

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<tr>
<td></td>
<td>Europe</td>
<td>Human + “human-like” (Britain)</td>
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</tbody>
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Reassortments among Influenza A Viruses Infecting Swine

Influenza A of Equines

- Equine influenza is an acute respiratory infection of horses, donkeys and mules
- Two distinct subtypes H3N8, and to lesser degree H7N7
- Clinical signs include pyrexia and a harsh dry cough followed by a mucopurulent nasal discharge; neurological signs have been described as a rare event
- Characteristically, influenza spreads rapidly in a susceptible population
- Interspecies transmission to dogs
Influenza A of Humans

• **Seasonal**
  - Established, widely circulating subtypes
  - Historically have included only H1, H2, H3, N1, N2
  - Currently H1N1, H3N2, and H1N2

• **Pandemic**
  - Influenza virus to which a majority of the human population has no existing immunity
  - Many influenza viruses circulating in animal species would meet this criteria
  - Emerges in the human population and efficiently transmits from human to human.
Background Influenza A in Humans

• Pandemics
  • 1918 “Spanish flu” (avian-like H1N1) - secondary infections
  • 1957 “Asian flu” (H2N2) – underlying conditions
  • 1968 “Hong Kong flu” (H3N2) – smoldering epidemic
  • 2009 “Pandemic H1N1” - ongoing

• Pseudopandemics
  • 1947: (H1N1) extreme intrasubtypic antigenic variation
  • 1976: “Potential Pandemic”, Fort Dix, New Jersey (“swine-like” H1N1)
  • 1977: Russian Flu, a Juvenile, Age-restricted Pandemic (“human-like” H1N1)
Influenza A – Molecular Epidemiology

- Ability to compare genetics of influenza viruses
- Compares the 8 genes to known, representative strains
- Fundamentally requires widely (publicly) available libraries with deposited genetic sequences
  - Reverse genetics of the 1918-19 pandemic virus
  - Clades of HPAI H5N1
  - Untangling the web between H1N1 (human seasonal) vs H1N1 (classical swine) vs H1N1 (avian-like, in swine) vs reassortments
Phylogeny of HA and NA Subtypes

Reverse Genetics

Clades of H5N1

Pandemic H1N1 2009

Gene Segments, Hosts, and Years of Introduction

- PB2, PA (~1998)
- PB1 (~1968)
- HA, NP, NS (~1918)
- NA, M (~1979)

Triple Reassortant
Classical Swine
Eurasian Swine

2009 A(H1N1)
Pandemic H1N1 2009

Garten et al. Science (2009); 325:197-201
Pandemic H1N1 2009

Garten et al. Science (2009); 325:197-201

2009 A(H1N1)
Highest Identity by BLAST
Human Infection Swine Virus
Human Seasonal

American Avian
Eurasian Swine

Classical Swine

A/Duck/NorthCarolina/91347/2001 H1N2
A/Swine/NorthCarolina/98225/2001 H1N2
A/Wisconsin/10/1998 H1N1
A/Swine/Minnesota/00194/2003 H1N2
A/Wisconsin/87/2005 H1N1
A/Ohio/01/2007 H1N1
A/Swine/Ohio/511445/2007 H1N1
A/Swine/Kansas/00246/2004 H1N2
A/Swine/Korea/ASAN04/2006 H1N2
A/Swine/Indiana/P12439/2000 H1N2
A/Swine/Guangxi/13/2006 H1N2
  A/Mexico/4115/2009
  A/NewYork/20/2009
  A/California/07/2009
  A/California/04/2009
A/PuertoRico/8/1934 H1N1
A/NewCaledonia/20/1999 H1N1
A/SolomonIslands/03/2006 H1N1
A/Brisbane/59/2007 H1N1
A/Duck/NewYork/13152-13/1994 H1N1
A/Swine/Saskatchewan/18789/2002 H1N1
A/Mallard/Maryland/161/2002 H1N1
  A/Mallard/Minnesota/SG-00121/2007 H1N1
A/Swine/Belgium/1/1983 H1N1
A/Swine/England/WVL7/1992 H1N1
A/Swine/Spain/50047/2003 H1N1
A/Swine/Zhejiang/1/2007 H1N1
Influenza A – Prevention and Control

C. Goldsmith, 1981
Basic Tenets

• Prevention
  • Biosecurity
  • Vaccination

• Control Options (depending on the situation)
  • Movement controls
  • Vaccination
  • Culling
Summary

- Influenza A viruses occur worldwide, affect humans and many other animals, and are very contagious.
- Immunity is subtype/strain specific – HA and NA.
- Antigenic shift and antigenic drift support development of new strains to which limited immunity exists in a particular population.
- Both classical and molecular epidemiology have provided great insight into influenza.
- Zoonotic transmission of influenza will play an ongoing role in influenza epidemiology.
Thank you for your attention

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