PANDEMIC INFLUENZA VIRUSES: PAST AND FUTURE

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NEW YORK

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THE BURDEN OF SEASONAL INFLUENZA

• 250,000 to 500,000 deaths globally/year

• More than 200,000 hospitalizations/year in US; deaths vary, more than 3,000 in 1986-7 and more than 48,000 in 2003-4

• $37.5 billion on economic costs/year in US related to influenza and pneumonia

• Ever-present threat of pandemic influenza

Sources: CDC, WHO, Am. Lung Assoc.
INFLUENZA VIRUSES CIRCULATING IN THE HUMAN POPULATION

Year

1918 1940 1960 1980 2000

H1N1
H2N2
H3N2
INFLUENZA VIRUSES CIRCULATING IN THE HUMAN POPULATION

- H1N1
- H3N2
- H2N2

Year:
- 1918
- 1940
- 1960
- 1980
- 2000

B

A

A

A

H1N1

H3N2

H2N2

H1N1
LIFE EXPECTANCY IN THE UNITED STATES 1900-2001:
BOTH SEXES

YEAR

AGE
NIH PROGRAM PROJECT GRANT

Mount Sinai School of Medicine
Adolfo Garcia-Sastre
Chris Basler
Peter Palese

CDC, Atlanta
Terrence M. Tumpey

University of Washington
Michael Katze

The Scripps Research Institute
Ian Wilson  Jim Paulson
REVERSE GENETICS

Viral RNA expression plasmids
- PB2
- PB1
- PA
- HA
- NP
- NA
- M
- NS

Protein expression plasmids
- PB2
- PB1
- PA
- NP

Transfection

Cells

Recombinant influenza virus
Characterization of the Reconstructed 1918 Spanish Influenza Pandemic Virus

Terrence M. Tumpey,1* Christopher F. Basler,2 Patricia V. Aguilar,2 Hui Zeng,1 Alicia Solórzano,2 David E. Swayne,4 Nancy J. Cox,1 Jacqueline M. Katz,1 Jeffery K. Taubenberger,3 Peter Palese,2 Adolfo García-Sastre2

The pandemic influenza virus of 1918–1919 killed an estimated 20 to 50 million people worldwide. With the recent availability of the complete 1918 influenza virus coding sequence, we used reverse genetics to generate an influenza virus bearing all eight gene segments of the pandemic virus to study the properties associated with its extraordinary virulence. In stark contrast to contemporary human influenza H1N1 viruses, the 1918 pandemic virus had the ability to replicate in the absence of trypsin, caused death in mice and embryonated chicken eggs, and displayed a high-growth phenotype in human bronchial epithelial cells. Moreover, the coordinated expression of the 1918 virus genes most certainly confers the unique high-virulence phenotype observed with this pandemic virus.

the HA from the Tx/91 virus with the remaining seven genes from the 1918 virus (Tx/91 HA:1918); a virus having the NA from 1918 with the remaining seven genes from the Tx/91 virus (1918 NA:Tx/91); and recombinant viruses having two 1918 (1918 HA/NA:Tx/91) or five 1918 genes (1918 HA/NA/M/NP/NS:Tx/91) with the remaining genes derived from the Tx/91 virus. The HA of the 1918 viruses used throughout these studies was derived from A/South Carolina/1/18 strain that was shown to preferentially bind the α2,6 sialic acid (human) cellular receptor (I6). The identity of the 1918 and Tx/91 influenza virus genes in the rescued viruses was confirmed by reverse transcription polymerase chain reaction and sequence analysis.

The infectivity of the 1918 virus and the ability to form plaques in the presence and in the absence of the protease trypsin were assayed in MDCK cells by the plaque method. The proteolytic cleavage of the HA molecule is a prerequisite for multicycle replication, and the ability of an influenza virus to replicate in the absence of trypsin has been thought to be an important factor...
Virulence of the 1918 virus in mice:
MLD50 log pfu

Texas/36/91

Tx/91: PB2, PB1, PA, NP, M, NS

1918: HA, NA

MLD50 log pfu

1918 “Spanish” flu

3.3

4.75
Virulence of the 1918 virus in chick embryos:
ELD50 log pfu

Texas/36/91  
Tx/91: PB2, PB1, PA, NP, M, NS
1918: HA, NA

1918 “Spanish” flu

1.5
Single gene reassortants identify a critical role for PB1, HA and NA in the high virulence of the 1918 pandemic influenza virus

Claudia Pappas, Patricia V. Aguilar, Christopher F. Basler, Alicia Solórzano, Hui Zeng, Lucy A. Perrone, Peter Palese, Adolfo García-Sastre, Jacqueline M. Katz, Terrence M. Tumpey

PNAS 105, 3064, 2008
## Virulence of 7:1 Reassortants (1918:Texas/91)

<table>
<thead>
<tr>
<th>Virus stock</th>
<th>LD$_{50}$</th>
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<tbody>
<tr>
<td>1918</td>
<td>3.25</td>
</tr>
<tr>
<td>1918:Tx PA</td>
<td>3.5</td>
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<tr>
<td>1918:Tx PB1</td>
<td>5.5</td>
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<tr>
<td>1918:Tx PB2</td>
<td>3.75</td>
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<tr>
<td>1918:Tx HA</td>
<td>&gt; 6</td>
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<tr>
<td>1918:Tx NP</td>
<td>3.5</td>
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<tr>
<td>1918:Tx NA</td>
<td>5.5</td>
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<tr>
<td>1918:Tx M</td>
<td>3.5</td>
</tr>
<tr>
<td>1918:Tx NS</td>
<td>3.25</td>
</tr>
<tr>
<td>Tx/91</td>
<td>&gt; 6</td>
</tr>
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</table>
Pandemic Influenza: What’s Next?
AVIAN INFLUENZA IS A THREAT
## Confirmed Human H5N1 Cases

**Updated August 31, 2010**

<table>
<thead>
<tr>
<th>Country</th>
<th>Cases</th>
<th>Deaths</th>
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<tr>
<td>Azerbaijan</td>
<td>8</td>
<td>5</td>
</tr>
<tr>
<td>Bangladesh</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Cambodia</td>
<td>10</td>
<td>8</td>
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<tr>
<td>China</td>
<td>39</td>
<td>26</td>
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<tr>
<td>Djibouti</td>
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<td>0</td>
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<tr>
<td>Egypt</td>
<td>112</td>
<td>36</td>
</tr>
<tr>
<td>Indonesia</td>
<td>168</td>
<td>139</td>
</tr>
<tr>
<td>Iraq</td>
<td>3</td>
<td>2</td>
</tr>
<tr>
<td>Lao</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>Myanmar</td>
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<td>Nigeria</td>
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<td>Pakistan</td>
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<td>Thailand</td>
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<td>17</td>
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<td>Turkey</td>
<td>12</td>
<td>4</td>
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<tr>
<td>Viet Nam</td>
<td>119</td>
<td>59</td>
</tr>
</tbody>
</table>

**Total** 505 300

WHO
INFLUENZA VIRUSES CIRCULATING IN THE HUMAN POPULATION

YEAR

1918 1940 1960 1980 2000

H1N1

H3N2

H2N2
Swine origin H1N1 to date

• First confirmed cases reported to WHO in late April 2009
• Global spread prompted WHO to declare pandemic 11 June 2009
• As of March 2010 the CDC estimates up to 80 million cases, as many as 362,000 hospitalizations and 14,460 H1N1-related deaths in the US
• 90% of hospitalizations and 88% of deaths occurred in individuals younger than 65 years of age
• On August 10, 2010, the WHO declares an end to the 2009 H1N1 pandemic
ORIGIN OF GENES OF THE 2009 SWINE H1N1 INFLUENZA VIRUS

North American avian (H1N1 or H3N2)

Human H3N2

Classic or Eurasian swine

H1N1 triple reassortant

Segments
1: PB2 - Avian
2: PB1 - Human
3: PA - Avian
4: HA – Swine (Classic)
5: NP – Swine (Classic)
6: NA – Swine (Eurasian)
7: M – Swine (Eurasian)
8: NS – Swine (Classic)
Number of Influenza-Associated Pediatric Deaths by Week of Death: 2006-07 season to present
Amino Acid Differences (%) in influenza A Virus Hemagglutinin (HA1)

A/South Carolina/1918

- A/New Jersey/1976
  - 19%
  - A/California/04/2009 PANDEMIC
- A/USSR/90/1977
  - 29%
  - A/Brisbane/59/2007 SEASONAL
THE 2009 SWINE H1N1 INFLUENZA VIRUS:

• TRANSMITS WELL
• HAS H1 (HEMAGGLUTININ) AND N1 (NEURAMINIDASE) SURFACE GLYCOPROTEINS SUGGESTING THAT THE HUMAN POPULATION HAS PARTIAL HERD IMMUNITY.
• DOES NOT EXPRESS THE VIRULENCE GENE, PB1-F2.
• IS SENSITIVE TO NEURAMINIDASE INHIBITORS.
The Guinea Pig as a Transmission Model for Influenza

- Anice Lowen
- Samira Mubareka
- Terry Tumpey (CDC)
- Adolfo García-Sastre
- Peter Palese
THE EPIDEMIC RESPIRATORY INFECTION AT CAMP CODY, N. M.

FREDERICK H. LAMB, M.D. (DAVENPORT, IOWA)
Captain, M. C., U. S. Army; Chief of Laboratory Service,
AND
EDWARD B. BRANNIN, M.D. (DALLAS, TEXAS)
First Lieutenant, M. C., U. S. Army; Chief of Pneumonia
Section, No. 1
CAMP CODY, DEMING, N. M.

Sept. 24, 1918, there was admitted to one of the
general medical wards of this hospital a soldier who
had just come from Camp Dix, N. J., with prisoners. He gave a history of having been sick for three
diagnosed influenza, and from the standpoint of this
report, it closed, Dec. 1, 1918. During this period
there were admitted to the hospital 3,265 cases; the
day of the greatest number of admissions was Octo-
ber 26, when 378 patients came in. October 30, the
hospital, though its normal capacity is 1,200 beds, was
accommodating 2,153 patients, of whom 1,899 had
influenza or pneumonia. In 624 instances, or 19 per
cent. of the 3,265, pneumonia was diagnosed. Death
occurred in 7.3 per cent. of the total number of cases
admitted, and in 38.4 per cent. of the pneumonias.
Two hundred and forty deaths was the toll during
the period mentioned.

At the beginning of the epidemic there were 4,239
officers and men in the camp. Of this number 637

Lamb, F.H., and Brannin, E.B. (1919). The epidemic
respiratory infection at Camp Cody. JAMA, April 12,
1056.
Transmission cage

Environmental chamber
Transmission of A/California/04/2009 virus (S-OIV) by the aerosol route

4/4 exposed guinea pigs were infected
Transmission of conventional swine and human influenza viruses

A/Swine/Texas/1998

A/Panama/2007/99

Nasal wash titer (log_{10} PFU/ml)

Day post-inoculation

1/4 infected

4/4 infected
Aerosol transmission of influenza virus from guinea pig to guinea pig is most efficient at low temperature.
TRANSMISSION SUMMARY

• THE GUINEA PIG TRANSMISSION MODEL WAS DEVELOPED
• THE H5N1 AVIAN INFLUENZA VIRUS DOES NOT TRANSMIT IN THIS SYSTEM
• AEROSOL TRANSMISSION OF INFLUENZA VIRUSES IS MOST EFFECTIVE AT LOW TEMPERATURE AND LOW RELATIVE HUMIDITY – PARTIAL EXPLANATION FOR SEASONALITY
VACCINES

KILLED (INACTIVATED)

LIVE (ATTENUATED)
TRIVALENT INFLUENZA VIRUS VACCINE

2009-2010

A/BRISBANE/10/2007 (H3N2)
A/BRISBANE/59/2007 (H1N1)
B/BRISBANE/60/2008
MONOVALENT INFLUENZA VIRUS VACCINE
(PANDEMIC H1N1, NOVEL H1N1, SWINE-ORIGIN)

2009/2010

A/CALIFORNIA/7/2009 (H1N1)
Percentage of Visits for ILI and H1N1 Vaccine Distribution, Sep 2009 – May 2010

Source: CDC ILI and Vaccine Distribution Data
TRIVALENT INFLUENZA VIRUS VACCINE

2010-2011

A/CALIFORNIA/7/2009 (H1N1)
A/PERTH/16/2009 (H3N2)
B/BRISBANE/60/2008
ANTIVIRAL RESISTANCE OF INFLUENZA VIRUS SUBTYPES

<table>
<thead>
<tr>
<th></th>
<th>M2 INHIBITORS (Adamantanes)</th>
<th>NA INHIBITORS (Oseltamivir)</th>
</tr>
</thead>
<tbody>
<tr>
<td>SEASONAL B</td>
<td>N/A</td>
<td>0%</td>
</tr>
<tr>
<td>SEASONAL A/H3N2</td>
<td>92%</td>
<td>0%</td>
</tr>
<tr>
<td>SEASONAL A/H1N1</td>
<td>25%</td>
<td>0%</td>
</tr>
<tr>
<td>PANDEMIC A/H1N1</td>
<td>100%</td>
<td>0.5%</td>
</tr>
</tbody>
</table>

Centers for Disease Control and Prevention