Newcastle disease virus

Siba Samal
Virginia-Maryland Regional College of Veterinary Medicine
University of Maryland
College Park
Newcastle disease

• First identified in Newcastle in 1926
• Affects all species of birds
• Mortality up to 100% in chickens
• Widespread in Asia, Africa, Europe and South America
• In USA, sporadic Newcastle disease (ND) occur due to importation of infected birds.
• In 2002 ND outbreak in California, four million birds were depopulated and cost the U.S. billions of dollars in damage and lost trade.
• NDV is a threat to the U.S. poultry industry and is listed as a select agent.
Newcastle disease virus

- A member of the genus *Avulavirus* in the family *Paramyxoviridae*
- The genome is a single-stranded negative-sense RNA consisting of 15,186 nucleotides
- The genome contains six genes in the order of 3’-NP-P-M-F-HN-L-5’
- The virus is enveloped, roughly spherical, with a diameter around 100-500nm.
- F and HN proteins form the external envelope spikes
- V and W proteins are produced from the P gene by alternative mRNAs that are generated by RNA editing.
NDV natural isolates

- Based on severity of disease, NDV isolates are grouped into three pathotypes:
  - Lentogenic strains
    - Cause mild or inapparent respiratory disease
  - Mesogenic strains
    - Cause respiratory or nervous signs with moderate mortality
  - Velogenic strains
    - Cause severe intestinal and/or neurologic disease resulting in high mortality
      - Neurotropic
      - Viscerotropic
Examples of pathogenicity indices obtained for strains of Newcastle disease virus.

<table>
<thead>
<tr>
<th>Virus Strain</th>
<th>Pathotype</th>
<th>ICPI&lt;sup&gt;a&lt;/sup&gt;</th>
<th>IVPI&lt;sup&gt;b&lt;/sup&gt;</th>
<th>MDT&lt;sup&gt;c&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ulster 2C</td>
<td>Lentogenic</td>
<td>0.0</td>
<td>0.0</td>
<td>&gt;150</td>
</tr>
<tr>
<td>Queensland V4</td>
<td>Lentogenic</td>
<td>0.0</td>
<td>0.0</td>
<td>&gt;150</td>
</tr>
<tr>
<td>Hitchner B1</td>
<td>Lentogenic</td>
<td>0.2</td>
<td>0.0</td>
<td>120</td>
</tr>
<tr>
<td>F</td>
<td>Lentogenic</td>
<td>0.25</td>
<td>0.0</td>
<td>119</td>
</tr>
<tr>
<td>LaSota</td>
<td>Lentogenic</td>
<td>0.4</td>
<td>0.0</td>
<td>103</td>
</tr>
<tr>
<td>H</td>
<td>Mesogenic</td>
<td>1.2</td>
<td>0.0</td>
<td>48</td>
</tr>
<tr>
<td>Mukteswar</td>
<td>Mesogenic</td>
<td>1.4</td>
<td>0.0</td>
<td>46</td>
</tr>
<tr>
<td>Roakin</td>
<td>Mesogenic</td>
<td>1.45</td>
<td>0.0</td>
<td>68</td>
</tr>
<tr>
<td>Beaudette C</td>
<td>Mesogenic</td>
<td>1.6</td>
<td>1.45</td>
<td>62</td>
</tr>
<tr>
<td>GB Texas</td>
<td>Velogenic</td>
<td>1.75</td>
<td>2.7</td>
<td>53</td>
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<tr>
<td>NY Parrot 70181 1972</td>
<td>Velogenic</td>
<td>1.8</td>
<td>2.6</td>
<td>51</td>
</tr>
<tr>
<td>Italien</td>
<td>Velogenic</td>
<td>1.85</td>
<td>2.8</td>
<td>50</td>
</tr>
<tr>
<td>Milano</td>
<td>Velogenic</td>
<td>1.9</td>
<td>2.8</td>
<td>50</td>
</tr>
<tr>
<td>Herts ’33/56</td>
<td>Velogenic</td>
<td>2.0</td>
<td>2.7</td>
<td>48</td>
</tr>
</tbody>
</table>

<sup>a</sup>ICPI, intracerebral pathogenicity index in day-old chicks  
<sup>b</sup>IVPI, intravenous pathogenicity index in 6-week-old chickens  
<sup>c</sup>MDT, mean death time (hr) for chicken embryos infected with one minimum lethal dose of virus  
(Adapted from *Diseases of Poultry*)
Relationship to human pathogens

- Closely related to human paramyxoviruses such as measles, mumps, human parainfluenza and respiratory syncytial virus, and the emerging pathogens, Nipah and Hendra viruses.

- Distantly related to lethal filoviruses, Ebola and Marburg, and the rhabdovirus, rabies.
NDV studies using reverse genetics

• Molecular Pathogenesis
  – Role of F, HN and V proteins in virulence
  – Glycosylation of HN and virulence

• Vaccine development
  – Improved Vaccines
  – Vaccine Vectors
Fusion Protein and Virulence

- rBeaudette C
  - NP
  - P
  - M
  - F
  - HN
  - L
  - Trypsin: -1.58
  - ICPI: 1.45
- rLaSota
  - NP
  - P
  - M
  - F
  - HN
  - L
  - Trypsin: 0.00
  - ICPI: 0.00
- LaSota V.F.
  - NP
  - P
  - M
  - F
  - HN
  - L
  - Trypsin: -1.12
  - ICPI: 0.00
HN Protein and Virulence

<table>
<thead>
<tr>
<th>Leader</th>
<th>rLaSota</th>
<th>rBC</th>
<th>rLaSo BCHN</th>
<th>rBC LaSo HN</th>
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<tbody>
<tr>
<td>MluI</td>
<td>NP</td>
<td>NP</td>
<td>NP</td>
<td>NP</td>
</tr>
<tr>
<td>AgeI</td>
<td>P</td>
<td>M</td>
<td>F</td>
<td>P</td>
</tr>
<tr>
<td>HN</td>
<td>M</td>
<td>F</td>
<td>M</td>
<td>M</td>
</tr>
<tr>
<td>L</td>
<td>F</td>
<td>HN</td>
<td>F</td>
<td>HN</td>
</tr>
<tr>
<td>Trailers</td>
<td>L</td>
<td>L</td>
<td>L</td>
<td>L</td>
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<tr>
<td>ICPI</td>
<td>0.29</td>
<td>1.58</td>
<td>0.75</td>
<td>1.02</td>
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<td>IVPI</td>
<td>0</td>
<td>1.45</td>
<td>0.38</td>
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</table>
V Protein and Virulence

**RNA editing - P gene**

---AAA AAG GGC CTA TGG TCG AGC---

**rBC**

---AAA AAG GGC CTA TGG TCt AG---

V (+1 frameshift)  
W (+2 frameshift)

**rBC/V-Stop**

---AAA AAG GGC CTA TGG TCt AG---

Truncated V

**rBC/Edit**

---AAg AAa GGC CTA TGG TCG AGC---

ICPI 1.58  1.45
IVPI 0.68  0

ICPI 0.72  0
HN protein glycosylation and Virulence

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<thead>
<tr>
<th></th>
<th>G1</th>
<th>G2</th>
<th>G3</th>
<th>G4</th>
<th>ICPI</th>
<th>IVPI</th>
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<tbody>
<tr>
<td>HN wt</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>1.50</td>
<td>1.32</td>
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<tr>
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<tr>
<td>HN G2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.87</td>
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<td>HN G3</td>
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<td></td>
<td></td>
<td>1.07</td>
<td>0.75</td>
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<td>HN G4</td>
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<td></td>
<td></td>
<td></td>
<td>0.66</td>
<td>0.31</td>
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<td>HN G12</td>
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<td>0.76</td>
<td>0.67</td>
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Newcastle Disease Virus as a Vaccine Vector for Infectious Bursal Disease Virus (IBDV)

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<tr>
<th></th>
<th>5'</th>
<th>100</th>
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<td>NDV</td>
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<tr>
<td>IBDV</td>
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</table>

% Protection

rLaSota

<table>
<thead>
<tr>
<th>3'</th>
<th>NP</th>
<th>P</th>
<th>M</th>
<th>F</th>
<th>HN</th>
<th>L</th>
<th>5'</th>
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<tbody>
<tr>
<td>VP2</td>
<td></td>
<td></td>
<td></td>
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<td></td>
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</tr>
</tbody>
</table>

rLaSota/VP2

<table>
<thead>
<tr>
<th>3'</th>
<th>NP</th>
<th>P</th>
<th>M</th>
<th>F</th>
<th>HN</th>
<th>L</th>
<th>5'</th>
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<tbody>
<tr>
<td>VP2</td>
<td></td>
<td></td>
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Current studies

- Role of individual NDV proteins in pathogenesis
- NDV-vectored human vaccines
- NDV-vectored veterinary vaccines
- NDV as an oncolytic agent
Floor Plan of Bio-Safety Level 3 Facility at University of Maryland

A – Autoclave
BC – Bio Safety Cabinet
C – Centrifuge
F – Freezer
I – Isolator

K – Key chest
R – Refrigerator
S – Sink
sl – Small Isolator
Conclusion

Reverse genetics of NDV enabled

• Identification of molecular basis of pathogenicity (F, HN and V proteins)
• Development of improved vaccines
• Development of NDV-vectored vaccines