CANINE INFLUENZA VIRUS

M. E. Spindel, DVM, MS; G. A. Landolt, DVM, PhD, DACVIM
“The patient in the next bed is highly infectious. Thank God for these curtains.”
Webinar Outline

• Overview of CIV
• Pathophysiology
• Clinical Signs
• Diagnostics
• Infection Control
• Therapy
• Considerations for Adoption
• Public Health Considerations
What is “Kennel Cough”?  

• Canine “URI” or “CIRD” is a common problem in many types of facilities
What is “Kennel Cough”? 

• There are bacterial, viral and mixed etiologies
• Each can present similarly
• Outbreaks have proven widespread & costly
• Shelters need diagnostic and management strategies
• Canine Influenza Virus (CIV) is a new differential
What is “Kennel Cough”?

**Bacterial Diseases**
- *B. Bronchiseptica*
- *Mycoplasma spp.*
- Secondary infections

**Viral Diseases**
- CIV
- Distemper
- Parainfluenza
- Adenovirus II
- Corona

**Other**
- Parasites
- Fungal dz

**Animal/Environment**
- Stress
- Crowding
- Biosecurity practices
- Age/health status
How often do you see “kennel cough”?
History of Canine Influenza Virus

• First identified Spring 2004

• Unusual respiratory disease outbreak at a Florida track
History of Canine Influenza Virus

- Twenty-two ill dogs
- 2 clinical syndromes
  - 14 dogs milder illness
  - 8 dogs peracute death w/ hemorrhage in respiratory tract
- Influenza virus recovered from one dog’s lung tissues

1Science 21, October 2005, Vol 310 No 5747, 482-485
Please click on your state if you think you have seen ClV.
What is Influenza?
Influenza in the News

“Pandemic”

“Horse flu epidemic”

Canine influenza virus surfaces
Researchers identify transmission of equine influenza virus to dogs

“Dog flu”

Everything you need to know about bird flu & what you can do to prepare for it
Jo Revill

Includes a comprehensive Q&A section
Influenza A Virus

Orthomyxoviridae

Two major envelope proteins

Hemagglutinin (HA) H1-H16

Neuraminidase (NA) N1-N9

e.g. H5N1, H3N2, H3N8

≠ Parainfluenza

Enveloped virus

Very easily inactivated with detergents and alcohols
Influenza A Virus Host Range

“Partial restriction of host range”
Avian-to-Human Transmission

Documented Avian Influenza infections in humans

Canada 2004
2 cases

U.S. 2002-03
2 cases

U.K. 2006
1 case

U.K. 2007
20 cases

Turkey 2006
12 cases
4 deaths

Thailand 2004-06
25 cases
17 deaths

Vietnam 2003-05
93 cases
42 deaths

Indonesia 2005-07
98 cases
78 deaths

Nigeria 2006
1 case

Egypt 2006-07
34 cases
14 deaths

Cambodia 2005-07
7 cases
7 deaths

Iraq 2006
3 cases
2 deaths

China 2003-07
25 cases
15 deaths

Netherlands 2003
89 cases
1 death

U.K. 2003
18 cases
6 deaths
2003
2 cases
1 death

Iraq 2006
3 cases
2 deaths

Hong Kong 1999
2 cases
2003
1 case

Hong Kong 1997
18 cases
6 deaths
2003
2 cases
1 death

Vietnam 2003-05
93 cases
42 deaths

Data as of 31 May 2007; http://europa.eu.int/comm/health
Cross-Species Transmission of Flu

“Bird Flu”

Virus maintained in “regular” host species

Virus NOT maintained in new host species

No human-to-human transmission
The Notable Exception

Canine influenza virus surfaces

Researchers identify transmission of equine influenza virus to dogs.

Canine influenza virus surfaces - 2004
Where Did the Virus Come From?

The flu virus found in greyhounds in Florida resembles the H3N8 virus found in horses.
Canine Influenza Facts

Greyhounds
CIV first isolated in 2004
Rapid spread in greyhounds across the United States
Crawford et al, 2006

Pet dog population
By winter of 2005, documented in humane shelters, in boarding facilities, and veterinary clinics
CDC, 2005
Canine Influenza Facts

- Spread among dogs in the United States
- Evidence of infection of dogs from >25 States
- Indicates that virus is being maintained in the dog population of the United States

In contrast... the UK experience
The UK Experience

Two separate occasions

Immunohistochemical and serological evidence in Sept 2002
Daly, 2005

Serological evidence in spring 2003
Newton et al., 2007
Virus Maintenance in Dogs

No evidence for virus maintenance in dogs in the UK.

Why was the virus maintained in dogs in the United States???
Why Are Some Viruses Maintained?

We really don’t know!
Canine Influenza Facts

• Appears that canine flu is “here to stay” in the U.S.

• We don’t know enough to explain the transmission of flu virus from horses to dogs.

• Much left to learn…
What Happens After Flu Infection?
What Happens After Flu Infection?

Nasal passages
Larynx
Trachea
Bronchus
Lung

Influenza replicates throughout the entire respiratory tract
What Happens After Flu Infection?

Inhalation of infectious virus

Infection and replication in the respiratory epithelium

Following release from cell, virus spreads quickly throughout the respiratory tract
What Happens After Flu Infection?

Efficient replication of virus in upper airway

Allows for efficient spread of virus by aerosol (up to 50 yards!)
Why Do We See Bacterial Infections?

Flu infected airway tissue
Airway cells are damaged = loss of cilia function

Reduced airway clearance can result in bacterial bronchopneumonia
What Happens After Flu Infection?

In uncomplicated cases:

- Healing of airway epithelium begins ~ 3-5 days after onset of illness
- Resolution of airway damage may take up to 3 weeks!
Clinical Signs
Clinical Signs

Who is affected?

- CIV is not preventable
- Most dogs are naïve
- 100% of exposed dogs are infected

- All ages
- All health ranges
- All sectors of canine population
Clinical Signs

Individual dog

- FEVER
- Cough
- Nasal discharge
- Vomiting
- Sneezing
- Anorexia & lethargy
- Dehydration
- Tachypnea & dyspnea
Clinical Signs

Population

• Rapid spread
• Prevalence increase
• Vx history insignificant
• Sicker dogs
  – Fever, nasal discharge, lethargy, peracute pneumonia
• Not responding to typical therapies
Clinical Signs

• Mild signs
  – Subclinical
  – Persistent cough
  – Low grade fever
  – Purulent nasal discharge
    • Usually due to secondary bacterial infection

• Severe signs
  – High fever
    • (up to 106°F)
  – Anorexia
  – Respiratory trouble
  – Pneumonia
  – Some dogs die despite supportive care
Clinical Signs

How will you know its CIV?

• CIV can look JUST like other causes of URI
  – Distemper
  – CPIV
  – CAV-2

• Two dogs with CIV can look VASTLY different
  – Virus
  – Host response
  – Timing of infection
Clinical Signs

How will you know its ClV?
How To Diagnose Canine Influenza?
How To Diagnose Canine Influenza?

Antigen detection
- IFA/HRP
- ELISA
- PCR

Culture
- Virus isolation

Serological analysis
- Fast turnaround
  - Non-viable virus
- Fast turnaround
  - Cheap + easy
- Isolate

- No isolate
- No isolate, Timing, Retrospective
- Slow, Timing, Viable isolate
Timing of Sample Collection

- Antigen detection (PCR)
- Virus isolation
- Serology
Diagnostic Principle

It may be necessary to combine several diagnostic tools to identify the etiological agent accurately and rapidly.
Which Animal To Sample?

Sample sick dogs (varying stages) +/- healthy contacts

Collect all potential samples (serum, whole blood, swabs)
Immunologically “naïve” animals (eg. young animals)
Which Animal To Sample?

“Lady”
- 2 yr female
- T: 104.1
- Lethargy

“Sidney”
- 2 yr female
- T: 104
- Neighbor to “Lady”

“Aubrey”
- 8 yr female
- T: 100.3
- Healthy

“Elly”
- 2 yr female
- T: 101.7
- Mildly lethargic

“Fog”
- 10 wk puppy
- T: 106.3
- Nasal d/c

“Buddy”
- 5 yr male
- T: 99.9
- Sick 4 wks ago
Timing of Sample Collection

Days

0 1 2 3 4 5 6 7 8 9 10 11 12

Antigen detection (PCR)

Virus isolation

Serology

......
Which Animal for Antigen or Virus Detection?

- **“Lady”**
  - 2 yr female
  - T: 104.1
  - Lethargy

- **“Sidney”**
  - 2 yr female
  - T: 104
  - Neighbor to “Lady”

- **“Aubrey”**
  - 8 yr female
  - T: 100.3
  - Healthy

- **“Elly”**
  - 2 yr female
  - T: 101.7
  - Mildly lethargic

- **“Fog”**
  - 10 wk puppy
  - T: 106.3
  - Nasal d/c

- **“Buddy”**
  - 5 yr male
  - T: 99.9
  - Sick 4 wks ago
Which Animal for Antigen or Virus Detection?

- "Lady": Positive
- "Sidney": Positive
- "Aubrey": Negative
- "Elly": Positive, weak
- "Fog": Positive, strong
- "Buddy": Negative
Timing of Sample Collection

- Antigen detection (PCR)
- Virus isolation
- Serology
Which Animal for Antibody Detection?

“Lady”
- 2 yr female
- T: 104.1
- Lethargy

“Sidney”
- 2 yr female
- T: 104
- Neighbor to “Lady”

“Aubrey”
- 8 yr female
- T: 100.3
- Healthy

“Elly”
- 2 yr female
- T: 101.7
- Mildly lethargic

“Fog”
- 10 wk puppy
- T: 106.3
- Nasal d/c

“Buddy”
- 5 yr male
- T: 99.9
- Sick 4 wks ago
Which Animal for Antibody Detection?

- "Lady" negative
- "Sidney" negative
- "Aubrey" negative
- "Elly" negative
- "Fog" positive, weak
- "Buddy" positive, strong
Virus/Antigen Detection
Virus Isolation

- Crucial for vaccines and epidemiology
- Often less sensitive
- Slow turnaround time
- Requirements:
  - Appropriate timing
  - Viable virus
  - Appropriate sample handling (viral transport medium, keep cool)
Testing Supplies for Virus/Antigen Detection

Nasal/pharyngeal swab
- 6 inch, polyester-tipped

Pharyngeal swab
- Less sensitive
- Viral transport medium

• Collect as much of the nasal secretion as possible
Antigen or Nucleic Acid Detection

**IFA**
- Tissue section
- Tissue imprint
- Nasal scraping
- Tracheal wash

**ELISA**
- Nasal swab
- Nasal wash

**PCR**
- Nasal swab
- Nasal wash
- Whole blood
- Tissue section
- CSF etc.
Antigen or Nucleic Acid Detection

• Does not require viable virus
• Faster turnaround time

• Immunofluorescence
• ELISA-based assays
• PCR-based assays
  – Conventional PCR
  – Real-time PCR
Polymerase Chain Reaction (PCR)

- Found wide application in diagnostic laboratories
- Highly sensitive method
- In theory: Can detect a single copy of the target nucleic acid
- Contamination!
Canine Influenza Diagnostics

The Colorado situation?
Spring 2006 to 2007
105 nasal swab samples
Acutely ill dogs
Canine Influenza Diagnostics

Client owned, Humane shelters (CO, WY, NE)
Dogs with clinical signs of “kennel cough” (n=105)

![Bar chart showing the number of animals tested positive for Canine Influenza using different diagnostic methods: Real-time PCR, Directigen™, and Virus isolation. N=62 for Negative and N=43 for Positive.](image)
Limitations of PCR-based Assays

• False positive results
  – Contamination
• False negative results
  – PCR inhibitors
• Does not differentiate live from dead organisms
• Always interpret results in conjunction with clinical information!*
A Quick Word About PCR Panels

• PCR Panels
  – Multiplex or single PCR assays
  – Screening for multiple agents in one sample
  – Reduces sample amount required for testing

• May make sense in a shelter situation
  – Cost for running single assay vs PCR panel

• Always interpret results in conjunction with clinical info!
Antibody Detection
Antibody Detection

- Cost-effective and easy to perform
- Excellent screening tool
- Circumstantial evidence
- Indicates exposure to antigen
  - Natural infection
- No prove of active infection
- Convalescent sample
Antibody Detection

False positive: prior exposure

False negative:
Sample was taken too early (or late)
Immunosuppressed (or young) animal
Which Diagnostic Laboratory to Use?

• Large number of laboratories offer PCR-based diagnostic tests
  – Private labs
  – State-run/University-associated labs

• Limited regulation of PCR-based test
  – Diagnostic kits and testing for animal movement and disease eradication: USDA approval
  – Am Association Vet Lab Diagnosticians (AAVLD): promotes voluntary quality control
    www.aavld.org/mc/page.do?
Which Diagnostic Laboratory to Use?

- Familiarity with the lab and other tests offered
- Published results in peer-reviewed journals?
- Expert available to answer questions?
- Quality control?
- Does the result make sense?
## Sample Handling

<table>
<thead>
<tr>
<th>Sample</th>
<th>Specific instructions</th>
<th>Test performed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nasal swab</td>
<td>In viral transport medium, keep at 4°C</td>
<td>Virus isolation, Ag detection, PCR</td>
</tr>
<tr>
<td>Blood sample</td>
<td>Serum: can freeze at –20 °C</td>
<td>Serology</td>
</tr>
<tr>
<td>Tracheal wash</td>
<td>Keep at 4 °C</td>
<td>PCR, culture</td>
</tr>
<tr>
<td>Tissue sample</td>
<td>1-2 cm³ blocks of tissue, keep at 4 °C</td>
<td>Virus isolation, PCR, Ag detection</td>
</tr>
</tbody>
</table>
Sample Transport

- Contact the laboratory
  - Shipping method
  - Hours of receiving
- Do not ship on Friday!
- Samples for virus isolation must be sent cold
- Viral samples are considered hazardous
- Must comply with shipping guidelines
Infection Control
Why Do Shelters Have Illness?

Many Factors Contribute!
Pre-Outbreak Considerations

Minimizing CIV exposure

1. Risk assessment
2. Risk aversion
3. Monitoring
1. Risk Assessment

How can we assess risk?

- CIV is not preventable
- Risk of introduction is always present
- Shelters possess MANY risk factors for perpetuating disease
1. Risk Assessment

So what can we do?

- **Understand transmission**
- **Segregate** the most susceptible from the most likely to infect
- Aim for **early recognition**
1. Risk Assessment

How does CIV spread?

- Dog-dog
- Aerosol
- Fomite transmission
1. Risk Assessment

How does CIV spread?

- Incubation
  - 2-7 days from exposure to clinical signs
  - some animals show no clinical signs

- Initial shedding
  - Peak shedding occurs PRIOR TO SYMPTOMS

- Duration of shedding
  - Estimate 7-10 days – some individual variation
Pre-Outbreak Considerations

Minimizing CIV exposure

1. Risk assessment
2. Risk aversion
3. Monitoring
2. Risk Aversion

Create individualized standard operating procedures

- Intake of animals
- Routine health procedures
- General husbandry
- Animal handling & movement
- Biosecurity
2. Risk Aversion

**Segregation**

- New Intakes
- All-in, all out movement
- Symptomatic animals

• What works in one facility may not work in another…
2. Risk Aversion

- Vaccination
- Stress Reduction
- Airway Health
2. Risk Aversion

How do vaccines help?

• Reduce secondary infections
• Increase immunity
• Can help distinguish outbreaks

Shelter “core” vaccines
Da2PP
Bordetella bronchiseptica
AAHA 2006 standards
Vaccinate ALL
Vaccinate on intake
Follow guidelines for timing
2. Risk Aversion

- Stress reduction
  - Physical
  - Emotional
  - Environmental

- Airway health
  - Avoid airway irritants
  - Decrease noise
  - Improve air quality
Pre-Outbreak Considerations

Minimizing ClV exposure

1. Risk assessment
2. Risk aversion
3. Monitoring
3. Monitoring

Train shelter staff to recognize disease EARLY
• Single animals & population trends

• Entry examination
• Record keeping
  • Track trends and recognize changes
• Frequent recheck of population
• Symptomatic animal examination
• Efficient notification of veterinary authority
Outbreak Considerations

What to do when disease strikes....
Outbreak Considerations

Apply general biosecurity measures

1. Establish biosecurity perimeters
2. Stop ALL animal movement
3. Sample ASAP for diagnosis
4. Quarantine
5. Manage sick animals
Outbreak Considerations

**SECONDARY PERIMETER**
Disease free – Still At High Risk

**PRIMARY PERIMETER**
Exposed and Ill Animals
Outbreak Considerations

CIV Perimeters

- Isolate EXPOSED – not just symptomatic!!!
- Standard of care = isolate “sick” animals
- CIV has ALREADY spread if symptomatic
- Ideally additionally isolate symptomatic animals
- Realistic shelter = isolate entire wards until not infectious
- Consider consolidating to create “clean” area
Outbreak Considerations

Ideal...

Realistic...
Outbreak Considerations

Ways to create CIV biosecurity perimeters

1. Stop admitting new animals (14 days)
   - outbreak will end
2. Off site admission of naïve (14 days)
   - outbreak will end
3. Admit naïve into onsite secondary perimeter (?)
   - Physically distanced
   - Ideally separately ventilated
   - STRICT biosecurity required
Outbreak Considerations

Stop moving dogs

Anyone or anything that enters and leaves the primary perimeter has the potential to transmit disease to another dog.
Outbreak Considerations

Animal management rules

• Ideal
  Designate CIV staff & non-CIV staff

• Less ideal
  Handle ill animals last

• Post clear signs
• Dedicate supplies & equipment
• Barrier clothing
• Wash frequently – labile virus
• Use footbaths
Treatment
Treatment

Practical Considerations

• Shelter philosophy
• Resources
• Population
• Cost of treatment
• Staff time
• Route/frequency/stress of medicating
• Is treatment needed??
Antimicrobial Treatment

• Viruses cannot be treated with antibiotics

• When to use antibiotics
  – Fever
  – Colored nasal discharge
  – Productive cough
  – Pneumonia
Antimicrobial Treatment

Choosing & using antimicrobials

• Bacterial resistance is real
  Use appropriate doses
  Give at correct intervals
  Give full course
  Don’t switch courses
  Avoid “big guns”

• There is no one perfect CIV antimicrobial

• Broad spectrum coverage

• Culture & sensitivity are important shelter tools
  Pneumonia
  Unresolving cases
  Severe disease
Other Treatment

**SUPPORTIVE CARE!!**
- Nutrition
- Hydration
- Minimize stress
- Avoid airway irritation

**OTHER**
- Antivirals
- Steroids
- Antitussives
### Antiviral Drugs

<table>
<thead>
<tr>
<th>M2 ion channel blockers</th>
<th>Neuraminidase inhibitors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Amantadine</td>
<td>Zanamivir (Relenza®)</td>
</tr>
<tr>
<td>Rimantadine</td>
<td>Oseltamivir (Tamiflu®)</td>
</tr>
</tbody>
</table>

- Must be given early in the course of disease
- **NOT RECOMMENDED FOR CANINE FLU:**
  - Proof of efficacy
  - Cost
  - Availability
  - Resistance
Special Considerations
Considerations for Euthanasia

- Most dogs recover with treatment
- Early diagnosis may improve survival
- Individual dog welfare
- Population resources
  - Treatment cost
  - Ability to provide care
  - Quarantine capability
- Necropsy is valuable diagnostic
Considerations for Adoption

• Halting admissions or adoption not always feasible

• CIV may become a recurrent issue

• Animals may recover best in home environment

• Ways to continue intake & release
  • Hold dogs past infectious period
  • Counsel reclaiming/adopting/visiting parties
  • Advise local veterinarians, shelters, animal groups
  • Provide written educational materials

• Become a trusted community resource
Canine Flu – Implications for Humans?
At present time: Minimal zoonotic potential of canine influenza
Canine Flu – Implications for Humans?

What will happen in the future?
Influenza Viruses Change!

Get the Flu Shot!

The reasons are all around you.

Influenza can lead to serious complications and even death for people in high risk groups. Protect yourself and those around you.

Get the flu shot. Not the flu.

Canadian Coalition for Influenza Immunization

www.influenza.cpha.ca
We Must Stay Vigilant!

- Monitor the spread of the virus through the United States
- Continue to work together to improve diagnosis, treatment & prevention of canine flu
- Continue to isolate and characterize canine viruses
- Stay vigilant when working with sick dogs

Realize the impact of canine influenza on the welfare of dogs
Contact Information

• Dr. Gabriele A. Landolt  
  landoltg@colostate.edu

• Dr. Miranda E. Spindel  
  mirandas@aspca.org

Veterinary Medical Center  
Colorado State University  
300 West Drake Road  
Fort Collins, CO 80523  
(970) 221-4535