Potential role of live animal markets and the environment in human exposure to avian influenza viruses

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Objectives

• Review data on presence of H5 and other avian and animal strains in live-animal markets and other environments
• Potential role of markets in human exposure
• Data on virus survival in environment
• Data on where virus is being isolated during outbreaks and inter-outbreak periods
Live poultry markets: Not all the same

- Fixed cages / vs temporary?
- Unsold poultry held overnight?
- Cage materials?
- Market hygiene practices?
- Species of poultry sold

HK LPM during Dec 1997
H5N1: Chicken-19.5%; Geese 2.5%
Duck 2.4% (Shortridge et al. 1999)

Avian influenza viruses: HK LPM
HK Poultry markets: 1999-2001

Unpublished data
Surveillance of H5N1 viruses in apparently “HEALTHY” poultry in live poultry markets in mainland China

- Why is isolation rate in markets so high?
- Very different to the farm-level perceptions

**Li et al Nature 2004; 430: 209-213**

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Isolation rates: Depends where you look

**Bali 2005**

- Community survey
  - 9 districts; Backyard flocks, small farms
  - 1202 swabs: H5N1=0
- 3 live poultry markets
  - 2 of 3 markets had H5N1
  - 133 swabs: H5N1=9 (7%)

*K Santia, AA Gde Putra, G Samaan and others - unpublished data*
Live poultry markets maintain and amplify avian influenza viruses

Senne et al. 1992
Bulaga et al Avian Dis 2003; 47: 1169-76
Kung et al. Avian Dis. 2003; 47: (S3) 1037-41

Influenza viruses amplify in live poultry markets:
Impact of “rest day” in retail markets on H9N2 isolation rates in chicken

Kung N et al. Avian Dis. 2003; 47: (Suppl 3) 1037-41
Live poultry markets maintain, amplify AND DISSEMINATE avian influenza viruses

Farms → Wholesale poultry market → Retail poultry

Kung et al Emerg Infect Dis 2007
Senne et al 1992

H9N2 isolation rates in live poultry markets in Hong Kong: 1999-2006: Impact of interventions

Impact of rest days, removal of quail and turnover on H9N2 virus isolation rates

Poisson generalized linear model adjusted for temperature, humidity, number of poultry sold, market factors, market rest days

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<tr>
<th>Year</th>
<th>Baseline</th>
<th>1xRD</th>
<th>2xRD</th>
<th>1xRD-Quail</th>
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Adjusted relative risk: Virus isolation

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<td>Chicken</td>
<td>1</td>
<td>0.73</td>
<td>0.61</td>
<td>0.56</td>
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<td>Minor poultry</td>
<td>1</td>
<td>0.42*</td>
<td>0.40*</td>
<td>0.37*</td>
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* = p<0.05

Lau et al EID 2007: 13: 1340-7
Poultry markets a risk factor for H5N1 disease?

Case-control:
- Hong Kong 1997: Mounts et al JID 1999; 180: 505-8
  - 15 cases vs. 41 matched controls
  - Exposure to LPM: OR 4.5 (1.2-21.7), p=0.045
  - Consumption or poultry or undercooked poultry, working in poultry industry (nil): NS

Anecdotal:
  - Of 6 urban cases,
    - all visited LPM within 2 weeks of onset, 5 within 1 week of onset
    - Only 1 had any other exposure to poultry
    - None had exposure to poultry outbreaks, dead poultry, etc
  - But NO CONTROLS
  - 127 cases, 2% LPM exposure, 21% exposure unclear.

Survival of LPAI and HPAI viruses in water

Temperature and salinity effects survival times

Time needed to lose 90% of infectivity
- At 17°C, no salinity,
  - LPAI H5 or H7 viruses take 29-111 days
  - HPAI H5N1 viruses take 16-26 days
- At 28°C, no salinity,
  - LPAI H5 or H7 viruses take 4-20 days
  - HPAI H5N1 viruses take 4-5 days

H9N2 virus (3.3 log10 EID50/ml)
- Survival in poultry water troughs after use: 3 days
- Fresh tap water: 12 hours
  Leung et al EID 2007; 13: 1380-2
Survival of H5N1 virus in dry or wet feces

**H5N1/97**

Survival at 37°C in wet feces for 6 days compared to 2 days with H5N1/97

Webster RG: www.who.int/csr/disease/avian_influenza/labstudy_2004_10_29/en

**H5N1/2004**

Survival at 37°C in wet feces for 6 days compared to 2 days with H5N1/97

Webster RG: www.who.int/csr/disease/avian_influenza/labstudy_2004_10_29/en

Survival of LPAI on porous and non-porous surfaces

- Inoculum of $10^3$-$10^4$/ml
- **Non porous surfaces**: Steel, tiles, gumboot, tyre, plastic, feather, egg shell: virus remains detectable for 72 hrs
- Porous surfaces:
  - Wood: 2 days
  - Egg tray, cotton or polyester fabrics: <1 day

Tiwari et al Avian Dis 2006; 50: 284-287
Heavy environmental contamination during H5N1 outbreaks, Cambodia

- Environmental samples collected in households within 1km of outbreak (avian or human)
- 14/43 households had positive environmental samples.
- In 14 positive households: 27/77 samples H5 RT-PCR positive (but culture negative)
  - Wet feces 2/4; pond water plants 1/2;
  - Mud under animal cages 2/4; ponds 4/19; yard 1/3;
  - Soil swabs: beneath houses on stilts 3/7; poultry places 5/7; poultry ranging places 2/4; feathers of dead poultry 3/6.
- Control households negative (Vong – pers comm)

Vong et al EID 2008; 14: 1303-5.

Summary

Live poultry markets:
- Amplify virus activity
- One key place to do systematic surveillance
- Source of infection to humans
- Disseminates infection back to farms
- Management interventions can have dramatic effect on virus load in markets

Virus survival
Virus contamination
- In village / farm environment
- Markets??
Gaps in knowledge

In H5N1 endemic regions
- Live poultry markets
  - Longitudinal surveillance on virus detection
  - Effect of market type and poultry mix
  - Impact of market interventions on virus circulation
  - Role in disseminating virus back to farming system
- Environmental contamination and virus survival
  - Methods for efficient virus isolation from environmental samples
  - Village settings
  - LPM setting including contamination of surfaces and poultry carcasses
- LPM exposure as a risk factor for human disease: Case control studies

Real world

- Rural poultry farm, poultry die off started 4 days ago, owner suspected avian flu H5N1
- Called in for urgent help…..
- ……..poultry buyer
- Surviving poultry purchased, to be sent to…
- ……..live poultry markets
- And the dead poultry…….?
Any surprise that LMP are a hot-bed of infection?
The question is not why there is human infection,
Rather, it is why there is so little!

Thank you