# Characterization of pathogens in non-human fecal matter

Session 2: What do we know about the pathogenicity of non-human source microbes?

#### Nicholas J. Ashbolt (Ashbolt.Nick@epa.gov) U.S. EPA/ORD/Cincinnati

Disclaimer: This presentation does not necessarily reflect official U.S. EPA policy

State-of-the-Science: Fecal Source Identification and Associated Risk Assessment Tools SCCWRP, 3535 Harbor Blvd. Costa Mesa, CA Nov 28<sup>th</sup>, 2012



## **Key Points**

- Zoonotic pathogens, by definition are infectious in humans
  - Issues for rec water: prevalence & survival
  - Common problem: resolution from non-pathogens
  - -Gastrointestinal & other illness (sequelae)
- Fecal but also environmental pathogens
  - Vibrio spp. & Ps. aeruginosa via wound infections
    Mycobacterium avium (increased virulence via animals)



## **Zoonotic pathogens**

#### Zoonosis – pathogen from animal-to-human

A few are viral (e.g. HEV pigs, H5N1 birds, hantavirus), range of pathogenic bacterial & parasitic protozoa

#### Which animal groups of concern in rec water?

- Water birds (e.g. [H1N1], Campylobacter jejuni, Cryptosporidium meleagridis, Salmonella enterica [Giardia lamblia], microsporidia)
- Mammals: cats & sea otters (Toxoplasma gondii), rodents (Leptospira\* & hantavirus), dogs (Toxocara & Trichuris spp.), rabbits (Cryptosporidium cuniculus)
  - \* 35 cases/y in Hawaii, Katz et al. 2011 EID 17:221-6



## **Non-GI illness endpoints**

- Prime interest is gastrointestinal (GI) disease
  But other endpoints / sequelae possible, e.g.
- *E. coli* O157:H7 GI, but may also cause HUS (hemolytic uremic syndrome)
- Campylobacter jejuni causes reactive arthritis & Guillain Barré syndrome
- Leptospira spp. freshwater leptospirosis



## **Understanding (fecal) sources**

- Animal surveys / by season / risk periods
- qPCR for various pathogens / indicators
- Microbial Source Tracking (MST)
  - Bacteroidales targets, yet poorly developed for non-ruminants & birds (Catellicoccus ?)
  - Emerging use whole genome sequencing
- Chemical biomarkers to ID sources
  - Fecal sterols & other biomarkers are aids



# Californian seagull risk?

 Most epidemiology studies to date have lacked statistical power, nor were they designed to specifically investigate health relationships to fecal indicators as well as other fecal source mixes

6



Schoen & Ashbolt (2010) Environ. Sci. Tech. 44:2286-91

A small portion (8%) of the fecal sequences were associated to *C. lari* or *C. jejuni* (clades C-D), while 85% constituted a distinct clade (A) away from *C. coli*, suggesting a novel species

Californian seagull Campylobacter **16S rRNA gene** sequence (n=255) similarities (97-99%) Lu et al. (2011) AEM 77:5034-5039





# Sandhill crane (Grus canadensis) campylobacters

- Campylobacter spp. were detected in 39%, 25%, and 11% of DNA extracts of the Sandhill crane excreta, water, and sediments, respectively
- Most identified as *C. jejuni* by PCR



8

\_u et al. (2011) ASM2011, May 21-24 New Orleans, Louisiana



### Further research gaps Zoonotic pathogens & indicators ?

- Known pathogens (Sal, Campy, Ec O157 etc.):
   Risk attributions? (human~cattle>pig/poultry)
- Emerging pathogens & what indicators?
  - HEV, *T. gondii*, *P. aeruginosa,* from pigs, cats, dogs, sea otters ...
- Antibiotic-resistant bacteria/genes from animal feeding operations & environmental amplification

9



### **Conclusions: what next**

- Probably need multiple qPCR targets to ID a source/mix with high sensitivity & specificity
- Then correlate to actual fecal (& environmental) pathogens for Fate &Transport modeling
  - Linking pathogen densities/behavior with surrogates
    - Noting differential die-off (of CFU) of FIB by fecal source!
  - Emerging roles for omics-ID/chemical markers
  - Emergence of ARG and antibiotic-resistant pathogens
- Dose-response models for various sequelae
- <sup>10</sup> other than GI illness



### Acknowledgements

Mary Schoen, Jeff Soller, Jingrang Lu, Ian Struewing, Helen Buse, Jorge Santo Domingo, Randy Revetta

