One Health: What can we learn about medical mycology from animals?

- Many fungal infections of humans also occur in wildlife species or domestic animals
- Identification of disease in animals may be early warning of emergence of disease in humans
- Animals can be models of disease pathogenesis
- Companion animals owners expect medical care equivalent to human care
  - Opportunities for clinical trials

Major Fungal Infections: Dogs and Cats

- **Molds**
  - Aspergillus (D > C)
  - Hyalohyphomycoses (D > C)
    - Talaromyces, Paecilomyces
  - Phaeohyphomycoses (uncommon)
    - Exophiala, Cladothialaphora, Alternaria etc.
  - Zygomycosis (rare)
    - Rhizopus, Mucor etc.
  - Dermatophytosis (C > D)

- **Immunosuppressed dogs at risk**
  - Cyclosporin, azathioprine, leflunomide, combinations

**Yeasts**
- Cryptococcus (C > D)
- Sporothrix (C > D)
- Histoplasma (C > D)
- Blastomyces (D > C)
- Coccioidoides (D > C)
- Candida (D > C)
- Malassezia (D > C)
The geographic distribution of mycoses in animals parallels that in humans.

What can dogs and cats tell us about risk for fungal infections in humans?

- 69 dogs with blastomycosis over 3 years1
- Incidence 1,420:100,000 versus 101:100,000 in humans
- 21% of owners reported one or more canine or human cases at the address
- Three addresses had > 1 dog affected
- Same distribution along waterways

59 dogs with blastomycosis over 3 years
- Incidence 1,420:100,000 versus 101:100,000 in humans
- 21% of owners reported one or more canine or human cases at the address
- Three addresses had > 1 dog affected
- Same distribution along waterways

What can dogs and cats tell us about risk for blastomycosis in humans?

<table>
<thead>
<tr>
<th></th>
<th>Blastomycosis (n = 59)</th>
<th>Quill-injured (n = 64)</th>
<th>Vaccinated (n = 169)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age</td>
<td>3.5 ± 3.0</td>
<td>3.8 ± 2.9</td>
<td>5.3 ± 4.2*</td>
</tr>
<tr>
<td>Male sex</td>
<td>40 (68%)</td>
<td>35 (55%)</td>
<td>57 (34%)*</td>
</tr>
<tr>
<td>Mixed breed</td>
<td>12 (20%)</td>
<td>28 (44%)</td>
<td>46 (27%)</td>
</tr>
<tr>
<td>Residence within 400 m of waterway</td>
<td>55 (95%)</td>
<td>40 (63%)*</td>
<td>125 (74%)*</td>
</tr>
<tr>
<td>Swimming</td>
<td>27/55 (67%)</td>
<td>41/55 (75%)</td>
<td></td>
</tr>
<tr>
<td>Exposure to excavation</td>
<td>24/55 (44%)*</td>
<td>9/55 (16%)</td>
<td></td>
</tr>
<tr>
<td>Exposure to beaver structures</td>
<td>20/55 (36%)</td>
<td>13/43 (30%)</td>
<td></td>
</tr>
</tbody>
</table>

What can dogs and cats tell us about risk for cryptococcosis in humans?


What can dogs and cats tell us about risk for coccidioidomycosis in humans?

- Case-control study; 41 dogs seen at UC Davis VMTH with coccidioidomycosis and 79 control dogs with other diseases, 2005-2013
- Client telephone survey, detailed travel histories
- Logistic regression analysis

From Grayzel et al. Transboundary Emerg Inf Dis 2016;Jan 22

Residences and travel for dogs with and without coccidioidomycosis referred to the UC Davis VMTH

Coccidioidomycosis cases (red points) and controls (green points). Point size is proportional to travel history to Arizona or Central Valley.

Association between dog demographic and environmental variables and coccidioidomycosis in dogs in California

<table>
<thead>
<tr>
<th>Variable</th>
<th>Cases (n = 41)</th>
<th>Controls (n = 79)</th>
<th>Odds ratio (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean age (months)</td>
<td>64 ± 36</td>
<td>99 ± 44</td>
<td>0.3 (0.16 – 0.56)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Digging behavior</td>
<td>25 (61%)</td>
<td>20 (25%)</td>
<td>6.68 (2.47 – 18.06)</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Travel to Arizona or California central valley</td>
<td>27 (66%)</td>
<td>40 (51%)</td>
<td>4.42 (1.46 – 13.38)</td>
<td>0.005</td>
</tr>
<tr>
<td>Male sex</td>
<td>26 (63%)</td>
<td>38 (48%)</td>
<td>2.32 (0.89 – 6.07)</td>
<td>0.079</td>
</tr>
<tr>
<td>Camping trips</td>
<td>14 (34%)</td>
<td>32 (41%)</td>
<td>2.62 (0.87 – 7.86)</td>
<td>0.077</td>
</tr>
</tbody>
</table>

Not retained in multivariable model:
- Age of home, property size, proportion of property with exposed soil
- Amount of time spent outdoors, visits to dog parks, hiking, working and sporting activities

From Grayzel et al. Transboundary Emerg Inf Dis 2016;Jan 22

The spatial distribution of canine coccidioidomycosis is closely correlated with the human incidence rate during 2012

- Spearman’s rho = 0.419 (P = 0.001)
- Additional higher risk area in north central CA, including Sutter, Butte and Yuba counties

From Grayzel et al. Transboundary Emerg Inf Dis 2016;Jan 22
What can dogs and cats tell us about Aspergillus fumigatus infections in humans?

- Sinonasal aspergillosis
- Common in dogs
- Chronic nasal discharge, sneezing, depigmentation
- Cribriform plate destruction
- Diagnosis and treatment expensive
- Requires repeated topical clotrimazole therapy

Risk factors for sinonasal aspergillosis in dogs

- 250 dogs with sinonasal aspergillosis at the VMTH, 1990-2014
- Reference population 190,894 dogs matching the catchment area
- Spatial patterns
- Association with environmental and climatic factors using a multivariate logistic regression model

<table>
<thead>
<tr>
<th>Variable</th>
<th>Odds ratio (95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>High traffic density</td>
<td>1.7 (1.1 – 2.8)</td>
<td>0.03</td>
</tr>
<tr>
<td>Wind difference (mean wind in 2014 – mean wind in 1990)</td>
<td>1.3 (0.99 – 1.7)</td>
<td>0.06</td>
</tr>
<tr>
<td>Active composting sites</td>
<td>1.2 (1.0 – 1.4)</td>
<td>0.03</td>
</tr>
<tr>
<td>Temperature difference (2014 – 1990)</td>
<td>0.69 (0.52 – 0.92)</td>
<td>0.01</td>
</tr>
<tr>
<td>Agriculture</td>
<td>0.67 (0.52 – 0.88)</td>
<td>0.003</td>
</tr>
<tr>
<td>Wind difference*temperature difference</td>
<td>1.6 (1.1 – 2.3)</td>
<td>0.01</td>
</tr>
</tbody>
</table>

29 other environmental and climatic variables not retained in final model, e.g., soil type, soil moisture, pesticide use, ozone, precipitation, fire history, vegetation types
What can dogs and cats tell us about Aspergillus fumigatus infections in humans?

- Effect of pollution
  - Sinonasal defenses
- Increased wind areas: coastal and urban areas with more construction?
- A. fumigatus thrives in composting piles
- Protective effect of agriculture
  - Fungicide use?
- Implications for human respiratory infections

What can dogs and cats tell us about clinical aspects of fungal infections in humans?

- Cryptococcosis is common in cats and dogs in California
  - ‘Cryptococcus neoformans’ not distinguished from Cryptococcus gattii
- What molecular types circulate in California?
Serum Cryptococcal Antigen Titers: Cats vs. Dogs


Canine Cryptococcus neoformans VNI infection

Canine Cryptococcus gattii infection (VGIIa or VGIIb)

Images courtesy Dr. Stacey Hoffman
Feline *Cryptococcus gattii* infection (VGII or VGIII)


Genetic Relationships Between Cryptococcus Isolates from Dogs and Cats in North America


C. gattii Isolates from Dogs and Cats Have Wider Variations in Antifungal Susceptibility than C. neoformans Isolates

‘Samantha’ 5 yo FS DLH

- Nasal cryptococcosis
  - C. gattii VGIII
- Initial response to fluconazole followed by relapse

‘Samantha’ 5 yo FS DLH

- Isolate 1 (JS75)

<table>
<thead>
<tr>
<th>Drug</th>
<th>MIC 9/30/10</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-flucytosine</td>
<td>0.12</td>
</tr>
<tr>
<td>Amphotericin B</td>
<td>0.25</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>0.50</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>≤ 0.015</td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.015</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.015</td>
</tr>
</tbody>
</table>

Poor response to treatment with fluconazole, SC amphotericin B, then itraconazole, then posaconazole

‘Samantha’ 6 yo FS DLH

<table>
<thead>
<tr>
<th>Drug</th>
<th>JS75</th>
<th>JS97</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-flucytosine</td>
<td>0.12</td>
<td>2.00</td>
</tr>
<tr>
<td>Amphotericin B</td>
<td>0.25</td>
<td>0.50</td>
</tr>
<tr>
<td>Fluconazole</td>
<td>0.50</td>
<td>256</td>
</tr>
<tr>
<td>Itraconazole</td>
<td>≤ 0.015</td>
<td>0.12</td>
</tr>
<tr>
<td>Posaconazole</td>
<td>0.015</td>
<td>0.12</td>
</tr>
<tr>
<td>Voriconazole</td>
<td>0.015</td>
<td>0.12</td>
</tr>
</tbody>
</table>
‘Samantha’ 6 yo FS DLH

- Serial exposure of isolate #1 to drug-free media required 87 transfers to lose resistance
- Serial exposure of isolate #2 to fluconazole-containing media led to resistance in 10 transfers
- Both isolates isogenic based on MLST
- Upregulation of ERG11 and PDR11 genes identified
  - ERG11: Lanosterol 14-alpha demethylase
    - Target of azole antifungals
    - 1.9-fold copy number increase
  - PDR11: Drug efflux pump
    - 2.6-fold copy number increase


‘Tuthra’ 8 yo MC DSH

- 5 week history of reluctance to jump, intermittent head shaking and sneezing
- ‘Clenching’ the furniture
- Treated by rDVM with amoxicillin-clavulanate, enrofloxacin, then metronidazole and prednisone 10 mg q24h
- Then began panting, crying, twitching
- Treatment changed to clindamycin after 3 weeks
- Referred

19

‘Tuthra’ 8 yo MC DSH

- 5 week history of reluctance to jump, intermittent head shaking and sneezing
- ‘Clenching’ the furniture
- Treated by rDVM with amoxicillin-clavulanate, enrofloxacin, then metronidazole and prednisone 10 mg q24h
- Then began panting, crying, twitching
- Treatment changed to clindamycin after 3 weeks
- Referred

‘Tuthra’ 8 yo MC DSH

- LCAT 1:32,000
- Deoxycholate amphotericin B
- Fluconazole 50 mg q12h
- Prednisolone 0.75 mg/kg q24h
- E tube feeding

‘Tuthra’ 8 yo MC DSH
Day 6

‘Tuthra’ 8 yo MC DSH
3 months

Titer at 1 year: 1:20

What can dogs and cats tell us about genetic susceptibility to fungal infections in humans?
What can dogs and cats tell us about risk for fungal infections in humans?

- 1998 – 2010: 9/31 dogs with cryptococcosis were American Cocker Spaniels versus 2.5% of the VMTH population (OR = 16, P < 0.001)\(^1\)
- 2005 – 2013: vizslas, Dalmatians, weimaraners, greyhounds, English pointers, bull terriers, Brittany spaniels and boxers were at increased risk for coccidioidomycosis (P < 0.01)\(^1\)
- 1990 – 2007: 20/30 dogs with disseminated aspergillosis were German shepherd dogs versus 4.5% of the VMTH population (OR = 41, P < 0.0001)\(^3\)
  - 77% were female
  - Rhodesian ridgebacks

Genetic analysis of the susceptibility to disseminated aspergillosis in dogs

- German shepherd dogs, Rhodesian ridgebacks and vizslas
- Genome Wide Association Analysis
  - Illumina canine HD SNP arrays
  - 30 affected dogs and 20 controls for each breed

Acknowledgements

- University of California-Davis
  - Beatriz Martinez-Lopez
  - George Thompson 3rd
  - Angela Gell
  - Danko Serebrjak
  - Barbara Byrne
  - Jonathan Sear
  - Ryan Garcia
  - Demetres Kapsogeorgis
  - LeAnn Lindsey
  - Eileen Samitz
  - Marisol Megro
  - Lisa Mulhine
  - Lisa Singer
  - Samiran Tread
  - Patricia Walsh
  - VMTH Clinicians and Pathologists

- University of Sydney, Australia
  - Gemma Fracassi
  - Richard Malk
  - Wieland Meyer

- Center for Companion Animal Health, University of California-Davis