Similarities between species

- We do not only look alike
- Live in same domestic environment
- Developed together over the last 10,000 – 15,000 years
- Receive preventative and health care

Dog in US society

- 70-80,000,000 dogs in the United States
- ~37\% of US households have at least one dog
- 63.2\% of households consider their dog a family member
- Many dogs now living to middle and geriatric age

AVMA 2012 Pet Ownership and Demographics Sourcebook

Dogs as a model

- Inbreeding has led to many breed predilection of disease
  - Many of these seen in humans
  - Allows smaller number of individuals to identify
    - Less background noise
- Many dogs are geriatric allowing for natural development of disease of aging – such as cancer
  - ~45\% of dogs >6 years in the US
Comparative medicine

- After humans most diverse and known disease occurrence
- ~400 inherited disease in dogs with human counterparts have been identified
- Affected by many of the same infectious diseases
- Affected by similar cancers

The dog as a model?

- Important to distinguish between laboratory animals and pets
- Toxicity testing in a “Large Animal” model
- Very different than dogs with spontaneous disease
Its in the DNA

- Share 650 Mb of shared ancestral DNA with humans
- Have closer DNA and protein sequences with humans than mice

Dogs informing human disease

Genetic studies

ORIGINAL ARTICLE

Chromosomal Aberrations in Canine Gliomas Define Candidate Genes and Common Pathways in Dogs and Humans

Peter J. Dickinson, BVSc, PhD, Dan York, BA, PhD, Robert J. Higgins, BVSc, PhD, Richard A. LeCouteur, BVSc, PhD, Nikhil Joshi, MS, and Danika Bannasch, DVM, PhD

Cancer types similar between species

- Lymphomas (NHL) and Leukemias
- Multiple Myeloma
- Soft Tissue Sarcoma
- Osteosarcoma (up to 75 x more common than in humans)
- Some forms of mammary cancer
- Melanoma
- Brain tumors (meningioma, glial)
- Bladder tumors
What is missing in mouse models?

- Long latency periods
- Genomic instability
- Tumor heterogeneity
- Microenvironment heterogeneity
- Metastatic patterns
- Often young mice used
- Often lean mice used
- Often immunocompromised mice used
- Differences in the immune system

Tumor microenvironment

- Tumor cells
- Stromal cells
- Immune cells
- Vasculature

Canine immune systems are genetically and developmentally much more similar to humans than rodent models
- Eg. TLR8 non functional in rodent
  - Unlike humans and dogs
- Tumors develop in spontaneous dog model in the face of intact immune system
  - Chronic inflammation
- Complexity of tumor immunity and suppression replicated
  - CD8+ T cells, Tregs, NK cells, APCs, Dendritic cells etc

Anti-CD28 humanized monoclonal ab
- Designed to induce T-cells
- Rodent models
  - Expanded T-cells without acute inflammatory reactions
- Biological differences between species
Advantage of a mouse model

- Short gestation
- Small in size
- Can manipulate individual genes
  - Organism
  - Particular tissues
- Immunocompromised variants
  - Nude mouse (athymic – T-cell deficient)
  - NSG mice models (T cell, B cell, NK cells, complement absent, defective macrophages & Dendritic cells)

Advantages of dog models

- Spontaneous tumor development
- Similar tumor microenvironment
- Intact immune systems
- Larger Size
  - Development of medical procedures
    - Limb sparing surgeries
  - Radiotherapy Trials
  - New Device and Drug Delivery Trials
Can’t we just build a better mouse model?

- Orthotopic xenografts
  - Better simulation of natural tumor environment
- Genetically engineered mouse models (GEM)
  - Often lack tumor heterogeneity
- Patient derived xenograft models (PDX)
  - Immunocompromised, mouse stroma
  - Very good at discerning new mechanisms

Translational canine melanoma study

- Radiotherapy to create tumor antigens
- CpG
  - Prokaryotic DNA sequences
  - Potent immune stimulation via TLR9
- Indolamine 2,3-Dioxygenase (IDO)
  - Immunosuppressive Enzyme
  - Inhibited by 1-Methyl-D-tryptophan (1-MT)
**Hypothesis**

![Diagram of XRT, IDO, 1-MT, CpG interactions]

**Canine clinical trial: schema**

![Schema diagram with timeline and interventions]
### Clinical trial: Results

<table>
<thead>
<tr>
<th>Age</th>
<th>Sex</th>
<th>Breed</th>
<th>Primary Disease</th>
<th>Systemic Disease</th>
<th>Best Primary Response (WHO)</th>
<th>Best Systemic Response (irRC)</th>
<th>Survival (months)</th>
<th>Toxicity</th>
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</thead>
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<tr>
<td>10</td>
<td>Female</td>
<td>Husky</td>
<td>Melanoma - lingual</td>
<td>Lung</td>
<td>PR</td>
<td>Lung – irSD</td>
<td>9.2</td>
<td>Mucositis – Grade 2</td>
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<td>8</td>
<td>Female</td>
<td>Labrador</td>
<td>Soft Tissue Sarcoma – body wall</td>
<td>LN, Lung</td>
<td>PR</td>
<td>LN – irPR</td>
<td>Lung – irSD</td>
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<td>Male</td>
<td>Mixed</td>
<td>Melanoma-buccal</td>
<td>Lung</td>
<td>PR</td>
<td>Lung – irPD</td>
<td>3.2</td>
<td>Mucositis – Grade 1</td>
</tr>
<tr>
<td>12</td>
<td>Female</td>
<td>Terrier</td>
<td>Melanoma-buccal</td>
<td>Lung</td>
<td>CR</td>
<td>Lung – irCR</td>
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<td>Melanoma-maxillary</td>
<td>Lung</td>
<td>PR</td>
<td>Lung – irPR</td>
<td>6</td>
<td>Mucositis – Grade 1</td>
</tr>
</tbody>
</table>

Canine melanoma median survival after development of metastatic disease:
- This Study: 5.8 months (95% CI: 3.2-9.2 months)
- Previously published historical controls treated with radiotherapy alone: 2 months (95% CI: 1-4 months)

### Clinical trial: Patient 1

- Alaskan Husky with rapidly progressive metastatic melanoma

**Pre-Treatment**

![Pre-Treatment Image]

**Post RT/Immunotherapy**

![Post-Treatment Image]
Decreased Tregs Post Treatment

T reg levels (CD45+CD3+CD8-CD4+FoxP3+)

Day 0

Day 28
Tumor Microenvironment Response

mRNA Expression by qPCR

Questions?