Krista La Perle, DVM, PhD, Dipl. ACVP
Professor
Director, Comparative Pathology & Mouse Phenotyping Shared Resource

Pathology Of Laboratory Animals
CL Davis & SW Thompson Foundation
August 5th, 2019
Outline

• Validation & reproducibility
• Controls
• Approaches to evaluating mice
• Unique Anatomy
• Miscellaneous Diseases
• Infectious Diseases
• Neoplastic Diseases
• DIY Pathology
• References
Validation of Animal Models

- Interpret morphologic changes resulting from genomic alterations and/or experimental manipulations
- Normal anatomy, physiology and histology
- Background strain-/breed-, age-, and sex-related lesions
- Infectious pathogens
- Husbandry practices
- Correlate with disease in various species
- Different species, tissues, pathologic processes & research disciplines
- Comprehensive, comparative and mechanistic!
Sentinel Health Monitoring Program

- French “Sentinelle” meaning watchtower
- Detect infectious agents in vivaria without need to test all animals
  - Exposed to soiled bedding from all cages on the rack
  - Direct contact
  - Air exhaust sampling
- Tested for excluded pathogens that can affect biomedical research
- Institutions differ in mouse strains used, protocols employed and excluded agents


Preclinical Studies

- Characterization of new genetically engineered mice & correlation with human/animal disease
- Preclinical toxicity/safety studies
- Preclinical therapeutic efficacy studies for new chemotherapeutics & chemopreventives in xenograft & metastasis models
- Pathogenesis of infectious agents & new treatment strategies
- Correlation of ante mortem imaging with post mortem pathology
- Evaluation of new surgical techniques and devices

Etc., etc.
Reproducibility

- Lack of scientific rigor
- Low statistical power
- Analytical flexibility
- Publication bias
Controls: Compare & Contrast!

- Individual
- Herd Health Issue

3-5 or 10! per age, sex, experimental group
### Controls: Compare & Contrast!

<table>
<thead>
<tr>
<th>NEGATIVE</th>
<th>SHAM</th>
<th>POSITIVE</th>
<th>COMPARATIVE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Spared from all manipulation</td>
<td>Mimics surgical procedure or treatment</td>
<td>Known manipulation results in reproducible effect</td>
<td>Gold standard care/treatment</td>
</tr>
<tr>
<td>Strain/substrain-, age-, sex-, genotype-matched in same room, caging, etc.</td>
<td>Vehicle/diluent; laparotomy &amp; “handle” liver vs. hepatectomy</td>
<td>Phenobarbital → proliferative liver lesions</td>
<td>Vaccine or chemotherapy</td>
</tr>
</tbody>
</table>

- Consistent personnel & sample size
- Random allocation & blinding
2 weeks following irradiation, T cell depleted allogeneic bone marrow transplantation and tail vein infusion of mesenchymal stem cells (MSCs)

No MSCs (3/3)  MSCs (1/1)  Abrogated MSCs (2/3)

- Variable sample size
- No mock irradiated/ transplanted
N = 8 slides

N = 30 slides

N = 22 slides

N = 15 slides

- FCA
- Adenoma
- HCC
Pieces of liver varied in size/shape & number (6-21/slide!)
Pathologist Involvement Early!

- Understand research question(s)
- Animal model advice/guidance
- Input on experimental design
- Necropsy protocol
  - Clinical pathology
  - Weights
  - Unique tissue harvest considerations
  - Ancillary testing

Flush with fixative & roll!
Anatomic Pathology

- Complete necropsy on live animals
  - Clinical evaluation
  - Bloodwork
  - Body/organ weights
  - Gross photos
  - Histopathology on complete tissue sets
- Biopsy or slide evaluation of selected tissue(s)
- Embryo/placental evaluation
- Tiered blinded approach to evaluating slides
Necropsy & Tissue Sampling

- Randomization
  - Subset of animals from all groups
- Live animals → body/organ weights, blood/body fluids, measure/weigh and photograph lesions
- goRENI
- GLP “lite”

Kittel et al, Exp Toxicol Pathol 55:413-431, 2004
Clinical Pathology

- **Hematology**
  - Complete blood count
  - White blood cell differential
  - Automated, manual smear evaluation
  - EDTA whole blood: ~25μl
  - Polychromasia

- **Chemistry**
  - Complete panel
  - Liver, renal, & lipid panels
  - Electrolytes, proteins
  - Plasma or serum: variable μl

- Urine/fluid analysis

- Cytology
Blood Sample Collection

- Values vary by site
- Hemolysis - ruptured RBCs
  - Pulling too hard on the syringe plunger during aspiration
  - Forcing blood in a syringe through a needle while transferring to a collection vial
  - Test parameter artifacts

http://inova.org/

Lab Animal 34: 39-43, 2005

https://theodora.com/rodent_laboratory/blood_collection.html
Mouse Anatomy

aKNITomy by Emily Stoneking on Etsy!
Brown Fat

- Multilocular
- High in mitochondria
  - Uncoupling protein (UCP)-1
- Thermoregulation
- Interscapular → “hibernating gland”
- Other locations
Skin & Hair

- Adult body skin is relatively thin
  - ~1-2 layers
- Hair growth in cyclic waves from cranial to caudal
  - Mostly in telogen (resting)
  - ~10% in anagen (growth), about 1-2 wks
  - As growth wave passes, follicle enters telogen.
  - Hair doesn’t fall out, remains in follicle until it is pushed out by next wave.
Harderian Gland

Retro-orbital tubululoalveolar gland that secretes porphyrins
Modified Sebaceous Glands: Preputial/Clitoral

Comparative Anatomy and Histology: A Mouse, Rat and Human Atlas, 2017
Modified Sebaceous Glands: Zymbal’s

Experimental Toxicologic Pathology 55: 91-106, 2003 (RENI)
Mammary Glands

- Mouse:
  - 5 pairs
  - Absent in adult male
  - Lobuloalveolar unit

$^{99m}$TcO$_4^-$ Scintigraphy
Hematolymphoid System

• Hematopoiesis
  □ Bone marrow
    ✓ 90-96% cellular
    ✓ Myeloid:Erythroid – 0.2-2.8:1; x = 1.5:1
  □ Spleen
    ✓ White vs. “red” pulp
  □ Liver, adrenal, LN, etc.

• Thymus doesn’t completely involute
  □ Indistinct Hassall’s corpuscles

• Perivascular lymphocytic aggregates in multiple tissues
CT mast cells everywhere!
Hematolymphoid System
Respiratory System

- Right: cranial, middle, caudal, accessory lobes; Left: single lobe
- Extra-pulmonary bronchi only
- Pulmonary veins contain cardiac and smooth muscle
- No BALT
  - PV/PB lymphocytes
  - Induced with age
Salivary Glands – Sexual Dimorphism

Submandibular gland: Granular convoluted ducts
Gastrointestinal

- Teeth: 2(I1/1, C0/0, PM0/0, M3/3) = 16
- Stomach: Limiting ridge
- Neonatal SI villar enterocytes are vacuolated
- Paneth cells in SI have robust granules!
- **Interepithelial** vs. lamina proprial mucosal mast cells
  - Mast cell protease (MCP) 1 [IE] vs. MCP4 (CT + LP)
  - ↑ w/ helminths, food allergies, GvH, etc., etc.
- Short rectum → susceptible to prolapse!
Liver

- Binucleated cells
- Polyploidy → diploid, 2n (mouse = 40)
- Anisocytosis/anisokaryosis

Intranuclear cytoplasmic invaginations
Pancreas

- Islet numbers & size vary by location
- Zymogen content is informational!
Kidney – Sexual Dimorphism

Cuboidal parietal epithelium of Bowman’s capsule
Kidney

- Proximal: P1, P2, P3
- Long segment: short segment nephrons 3:1 → greater concentrating ability
- Medulla: outer (outer & inner stripes) & inner

Figure 1: Schematic drawing of the nephron and vasculature, denoting subtopographical anatomic relationships.

Figure 2: Subgross topographical anatomy of the rat kidney illustrating the zones of the kidney with correlative sites of functional activity and nephrotoxic examples targeting the zone or function in Table 1.
Urine

- Urinate when handled
- Small volumes
  - Pool vs. metabolic cages
- 1.060-1.080 specific gravity
- Proteinuria normally!
  - Higher in males vs. females
  - Major urinary proteins = α₂μ globulins
    - Potent human allergens
    - Pheromones
    - Mouse m 1
Male Reproductive

Os penis;
Copulatory plug;
Spermatogenesis = 35 days
Female Reproductive

**Table 1.** Basic classification of the stages of estrous cycle based on cell types and relative numbers of these cell types in vaginal smears.

<table>
<thead>
<tr>
<th>Stage</th>
<th>Neutrophils</th>
<th>Small nucleated epithelial cells</th>
<th>Large nucleated epithelial cells</th>
<th>Anucleated keratinized epithelial cells</th>
<th>Relative cell density</th>
</tr>
</thead>
<tbody>
<tr>
<td>Proestrus</td>
<td>0 to +</td>
<td>+ + to +++</td>
<td>0 to +</td>
<td>0 to +</td>
<td>Low to moderate</td>
</tr>
<tr>
<td>Estrus</td>
<td>0 to +</td>
<td>0 to ++</td>
<td>0 to ++</td>
<td>++ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Rat</td>
<td>0 to +</td>
<td>0 to +</td>
<td>++ to +++</td>
<td>++ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Mouse</td>
<td>0 to +</td>
<td>0 to +</td>
<td>++ to +++</td>
<td>++ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Metestrus</td>
<td>+ to +++</td>
<td>+ to ++</td>
<td>+ to ++</td>
<td>+ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Rat</td>
<td>+ to +++</td>
<td>+ to ++</td>
<td>+ to ++</td>
<td>+ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Mouse</td>
<td>+ to +++</td>
<td>0 to +</td>
<td>+ to ++</td>
<td>+ to +++</td>
<td>Moderate to high</td>
</tr>
<tr>
<td>Diestrus</td>
<td>++ to +++</td>
<td>+ to ++</td>
<td>+ to ++</td>
<td>0 to +</td>
<td>Low to moderate</td>
</tr>
</tbody>
</table>

*Note: 0 = none; + = few; ++ = moderate; +++ = high.*

**Toxicologic Pathology 43: 776-793, 2015 (rat and mouse vaginal cytology)**

Granulocytes ~*~ Hemosiderophages
Placenta

- 21 day gestation
- Discoid: single & shaped like a disc
- Labyrinthine: maternal-fetal interdigitation
- Hemo(tri)chorial: no maternal uterine layers retain; chorion bathed in fetal blood
Adrenal Gland

- Inapparent zona reticularis: no 17α hydroxylase
- X-zone +/- degeneration +/- ceroid/lipofuscin (mouse)
- Subcapsular spindle cells (mouse)
  - Reserve cells; spindled (type A) or polygonal lipid-laden (type B)
- Accessory cortical nodule
Nervous System

- Lissencephaly
- C7-T13-L6-S4-Cd28
Bone

- Cortical bone = circumferential lamellae (A) vs. Haversian systems (osteons) (B)
- Longitudinal growth ceases ~3-6 months
  - Incomplete physeal closure
Mouse Pathology

• Signalment
  - Strain, age, sex

• Husbandry
  - Caging/bedding, HVAC, diet

• Sentinel health status
  - Know what is on institution’s exclusion list
  - Positive diagnostic ≠ pathology
  - Housing location

• Experimental manipulation

• Institutional/geographical variability

• Communicate with clinical laboratory veterinarians & scientific staff!
Miscellaneous Diseases

HouseMouseOnCheese.com
Inbred Strains

- Product of $\geq 20$ consecutive generations of sister x brother or parent x offspring matings
- Single ancestral pair in the 20th or subsequent generation
- Homozygous at virtually all loci
- Strain-distinguishing characteristics
FVB/N

- Primarily used for...
  - Random transgenesis
  - Large pronuclei
  - Natural superovulators
  - Good mothers

Intramural.nihm.nih.gov/tgc/photogallery.html
Retinal Degeneration

• $Pde6b^{rd1}$
  - *Retinal degeneration 1* mutation
  - FVB, C3H, SJL, SWR, CBA, outbred swiss
  - Phosphodiesterase 6b, cGMP, rod receptor, beta polypeptide
  - Blindness when homozygous recessive
    - Model for retinitis pigmentosa
      - Nyctalopia (dim light/night) → reduced peripheral visual field
        ± loss of central vision
  - Transgene and $Pde6b^{rd1}$ segregate independently
Retinal Degeneration
Space Cadet Syndrome

- F>M
- Withdrawal from social interaction
- Seizures $\rightarrow$ hypoxia $\rightarrow$ neuronal necrosis
  - Spontaneous, excitotoxins or audiogenic (tattooing, clipping, noise, etc.)
  - Cerebral cortex, hippocampus, thalamus
  - Often no lesions!
  - ✓ Concurrent astrocyte hypertrophy and gliosis $\rightarrow$ chronicity?
  - ✓ Brain weight differences?
Space Cadet Syndrome
Hernias

- Lateral femoral hernias (FVB/NHsd; F>M)
- Scrotal hernias (FVB/N, closed colony, M only)
- Other causes
  - Muscle weakness
  - Estrogen in males; testosterone in females
  - Genotype (↓ fibulin 3; ↑ IGF-3)
Hernias

Lateral

Scrotal
Prostate Mucinous Metaplasia

**Comparative Medicine** 66: 286-289, 2016

<table>
<thead>
<tr>
<th>Table 1. Rate of mucinous metaplasia in FVB/N mice</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age of mice (mo)</td>
</tr>
<tr>
<td>No. of mice</td>
</tr>
<tr>
<td>% of mice with PAS-positive areas</td>
</tr>
<tr>
<td>Average no. of PAS-positive areas per mouse</td>
</tr>
<tr>
<td>% of lesions in each lobe</td>
</tr>
<tr>
<td>Laterality</td>
</tr>
<tr>
<td>Dorsal</td>
</tr>
<tr>
<td>Ventral</td>
</tr>
</tbody>
</table>
Primarily used for...

- Targeted mutagenesis by homologous recombination
  - Knock-outs, knock-ins
- Embryonic stem (ES) cell lines
- Numerous (P, S, T, X) substrains
Acidophilic Macrophage Pneumonia

- Eosinophilic crystalline pneumonia; crystalline pneumonitis
- NMRI, T x HT, 129, C57BL/6 & B6;129
- Asthma and parasitic models
- Inhalational toxicology studies
- Pulmonary tumors and pneumocystosis
- Up to 88% incidence
- Cause of death in up to 10%
- Associated with epithelial hyalinosis
Acidophilic Macrophage Pneumonia
Epithelial Hyalinosis

- Olfactory, Tracheal
- Bronchiolar
- Gastric
- Biliary, Pancreatic
- Ureteral
- Esophageal
Charcot-Leyden Crystals

- Ultrastructural resemblance
- Primates and NHPs
- Pathologies associated with eosinophils
  - Derived from core of eosinophil granules
- Hexagonal, bipyramidal
- High in Zn, lysophospholipase
- Form directly
AMP/Hyalinosis Crystals

- Iron
- $\alpha_1$-anti-trypsin
- Immunoglobulin
- Granulocyte breakdown products
- **Ym1**: Lung, spleen, BM
  - T lymphocyte-derived eosinophil chemotactic factor (ECF-L)
  - Azurophilic granules in neutrophils
- **Ym2**: Stomach, thymus, kidney
  - Precursor (55 kd; glandular) vs. mature (45 kd; forestomach)
- **AKA**: Chitinase 3-like 3 (Chil3) and Chil4 (marker of alternatively activated macrophages)
• Crystals form due to local $\uparrow$ 2° to neutrophil degranulation during repeated episodes of inflammation
  □ Also seen concurrently w/ neoplasms
• Th2 $\rightarrow$ macrophages (M2) $\rightarrow$ phagocytosis +/-rupture $\rightarrow$ IL1β, IFNγ, IL4, IL17
  □ Mildly acidic pH contributes to crystallization and phagolysosome formation
• Crystals resistant to degradation
• Inflammation, epithelial hyperplasia

https://www.brainpickings.org/2013/02/01/which-came-first-the-chicken-or-the-egg/
Corpus Callosum Hypo-/Aplasia


Slow, clumsy \(\rightarrow\) impacts behavioral testing (i.e. wheel running)
Megaesophagus

Normal

Abnormal

Impaction +/- morbidity
C57BL/6

- Primarily used for…
  - Blastocyst injection with ES cells
    ✓ Hardy
  - Respond well to superovulation
  - Mouse genome sequenced

Nature, 420, 2002
Hydrocephalus
Malocclusion

- Incisors: open roots, grow throughout life
- Ptyalism
- Cellulitis
- Weight loss
- Powdered diet, tooth trimming
Also due to senility, irradiation

Posterior migration of lens capsule epithelium, lens fiber degeneration/necrosis, bladder/balloon cells, Morgagnian globules

Cataracts & Microphthalmia/Anophthalmia
Corneal Opacities

• DDX
  - ↑ ammonia levels in cage
• Calcific band keratopathy: Ca & P w/ less Fe, Zn, Na, Al
  - Ketamine anesthesia
    - Attenuated blink response → tear film evaporation → exposure keratitis
  - Mechanical irritation
    - Nude mice
    - Rubbing eyes/face
    - Caging, waterers
Barbering, Dalila Effect, Trichotillomania
Ulcerative Dermatitis

- F > M, ↑ w/ age, peaks in spring/fall or midsummer
- Excoriations, alopecia → erosions, ulceration → fibrosis, contracture
- Pruritic!
- Head, neck, dorsum
- Lymphadenopathy, splenomegaly
  - Lymphoid hyperplasia, plasmacytosis, extramedullary hematopoiesis
- Opportunistic infections with *Staphylococcus* or *Streptococcus* spp.
- Cause?
  - Behavioral, immune complex vasculitis, cellular oxidative injury, vitamin A toxicity
  - Barbering, fur mite hypersensitivity, fight wounds, wire bar lids/waterers
  - High fat diets vs. calorie restriction
Ulcerative Dermatitis

• “Treatment”
  - Antibiotics (topical, feed, water)
  - NSAIDs
  - Vitamin E (gavage with Derm Caps, diet)
  - Topical cyclosporine, zinc oxide, povidone-iodine + sulfadiazine, *sodium hypochlorite*
  - Rx interferes w/ research → euthanize!

JAALAS 51: 586-593, 2012
Presbycusis

- Age-related hearing loss → may alter behavior
- Genetically complex (Cadherin 23 [Age-related hearing loss 1] mutation, among others)
- Disruption/loss of cochlear hair cells → ganglion degeneration → collapse of organ of Corti
- C57BL/6, 129, A/J, DBA, etc.

Gerontology 61: 149-157, 2015
Imperforate Vagina → Muco-/Hydrometra
Melanosis
Osteoarthritis
Cardiac Calcinosis

- BALB/c (esp. ByJ substrain), SCID, C3H, DBA
  - 90% vs. 3% calcinosis at 5 wks
  - 30% vs. 1% total surface area
  - Lymphocytes and macrophages
  - Translocation of connexin 43 from intercalated disk to sarcoplasm

- Dystrophic mineralization +/- fibrosis
- RV > LV, IVS, atria
  - BALB/c – epicardial
  - C3H – myocardial with degeneration
  - DBA – epicardial, myocardial, soft tissue
Cardiac Calcinosis

- Increased with exogenous corticosteroids or ACTH, high fat/low protein diets, and selenium/vitamin E deficient diets
- Genetics
  - Dyscalc1 locus on chromosome 7 in C3H and DBA
    - Possible candidate genes: Abcc6, EMP-3, Rcn-3, Kcnj14
- Normocalcemic
- Usually no clinical cardiac dysfunction
- Von Kossa, Alizarin red special stains
Non-Inbred Strains

- F1 Hybrids: generated by crossing 2 different inbred strains
- Outbred Stocks: intentionally not bred with siblings or close relatives to maintain maximum heterozygosity
Nasal Septal Eosinophilic Substance

- PAS w/ diastase positive
- Congo red negative
- Trichrome positive
- Collagen + complex CHO
True Hermaphroditism

- Most ES lines are XY
- Blastocysts for injection are XX or XY
- Both cell types have significant contributions to gonads
  - >30% XY cells \(\rightarrow\) males
  - <20% XY cells \(\rightarrow\) females
  - 20-25% XY cells \(\rightarrow\) hermaphrodites
    - Ovotestis/testis on left
    - Ovary on right
    - Cystic mullerian duct remnants
- Pseudo: \(2^0\) sex characteristics differ from gonadal tissue (name based on gonad)
True Hermaphroditism
Fibro-osseous Lesions

- B6C3F1 mice
- Tibia, femur, sternum, vertebrae, nasal bone
- No renal/PTG changes
- Females +/- cystic ovaries/endometrial hyperplasia
- ↑ w/ DES (estrogen) & misoprostol (PGE1 analogue)
Lesions Lacking Strain Specificity

http://www.britam.org/DNA/BAMAD82.html
Mouse Urologic Syndrome

- Males
- Retention of ejaculated coagulum in urethra
- Found dead
- Urine dribbling, ulcerative balanoposthitis, paraphimosis, urethritis, prostatitis, cystitis
- Damage to external genitalia via fighting or self-induced trauma
- Seizures
Amyloidosis

• Secondary, reactive: AA
  - Chronic inflammation
  - Macrophage activation $\rightarrow$ IL-1, IL-6, TNFα $\rightarrow$ hepatic production $\rightarrow$ proteolysis
  - Spleen, liver, SI, kidney (glomeruli), heart

• Primary, senile: AapoAII
  - Hepatic production $\rightarrow$ no proteolysis
  - Intestines, endocrine and reproductive organs, less severe in spleen and liver

• Deposits are typically mixed
Amyloidosis
Other Urinary Disorders

- Chronic glomerulonephropathy
  - Membranoproliferative
  - PAS positive, immune-mediated

- Chronic progressive nephropathy
  - Multifocal: glomerular hyalinosis, glomerulosclerosis, tubular degeneration and regeneration, interstitial inflammation, hyaline casts

- Hydronephrosis/-ureter
  - Unilateral to bilateral

- Tubular necrosis
  - Chloroform fumes: adult male DBA, C3H mice, other genotypes
  - NSAIDS (banamine): at C:M junction with mineralization
Urolithiasis

- Estrogen supplementation in immunodeficient w/ mammary xenografts (release pellets)
  - ↑ urethral resistance → urethral occlusion → urinary retention
  - Cystitis, pyelonephritis, urolithiasis
  - Struvite calculi from urease-producing bacteria:
    - *Proteus mirabilis*
    - *E. coli*
    - *Staph intermedius, S. xylosus*
Anasarca

- Amyloidosis
- Glomerulonephropathy
Auricular Thrombosis

- Left- or right- sided heart failure
  - Common cause of non-infectious dyspnea
- Left auricle
- ± Glomerular amyloidosis
  - $\rightarrow$ antithrombin III
  - Also in BALB/c which are resistance to amyloid
Necrotizing Polyarteritis

- Murine counterpart to rat/NHP polyarteritis nodosa and K9 beagle pain syndrome/juvenile polyarteritis
- Small- to medium-sized arteries
  - Not lung!
- Fibrinoid degeneration, neutrophilic to lymphoplasmacytic inflammation, myointimal hyperplasia and fibrosis
- Segmental, acute to chronic, multiple arteries
- Immune complexes
  - Cause?
- Vestibular syndrome: circling, rolling
- DDX: Spontaneous aortitis (root) in Balb/c mice
  - *Toxicologic Pathology* 37: 667-671, 2009
Necrotizing Polyarteritis
Brainstem Infarction

- Swiss mice, females, <1-year-old
- Caudal cerebellar artery (thin arrows)
  - Branch of basilar artery (*) that emerges distal to convergence of vertebral arteries (thick arrows)
- **Veterinary Pathology 48: 726-729, 2011**
- Vestibular syndrome
CNS Epithelial Inclusion Cyst

- Common, especially spinal cord
- Typically occur on midline, and although
- No clinical signs
- Also called epidermoid cysts
  - Skin
  - Ovary
Cystic Endometrial Hyperplasia

+/- Klebsiella oxytoca pyometra

NTP Nonneoplastic Atlas
Cystic Bulbourethral Glands

Fertility

+/- Suppuration w/ bacteria (Staph, Pasteurella pneumotropica)
Seminal Vesicular Dilation or Atrophy
Preputial Gland Acinar Atrophy & Ductal Ectasia

Normal

Senile Atrophy
Ringtail / Annular Constrictions

- Prolonged vasoconstriction $\rightarrow$ dry gangrene, necrosis, auto-amputation
- Multifactorial
  - Genetics (epidermal differentiation genes)
  - $\downarrow$ relative humidity (25-40%; winter, early spring)
  - $\uparrow$ ambient temperature (>80F)
  - Hydration status/nutrition
- DDX
  - Frostbite, mouse pox, *Staph/Strep* dermatitis, suckling mice and cotton nestlets
Disordered cornification $\rightarrow$ hyperkeratosis, abnormal desquamation $\rightarrow$ persistent keratin ring $\rightarrow$ strangulation

*Veterinary Pathology* 52: 700-711, 2015

**Ringtail / Annular Constrictions**
Auricular Chondritis

- Ear tags
- Tagged and untagged
- Copper and iron released from tag → ↑ reactive oxygen species → inflammation, fibrosis, brittle cartilage
- ↑ metallothionase (MT) I and II
- Result autoimmune processes

*Arthritis Research & Therapy* 8:R134, 2006
Periodontitis (Hair Tooth) & Glossitis (Hair Tongue?!)
Bacterial Diseases

http://www.ehow.com/about_5380594_bacteria-life-cycle.html
Citrobacter rodentium

- Gram negative bacilli
- Transmissible murine colonic hyperplasia
  - Hyperplastic colitis
- Direct contact, fecal-oral
- Transient SI colonization
- Attachment to descending colon
  - Bacterial intimin and translocated intimin receptor (Tir)
    - Dissolution of brush border, actin rearrangement, pedestal formation
    - Like attaching and effacing EPEC & EHEC → model!
Citrobacter rodentium

- Bacterial colonization elicits mucosal hyperplasia
  - Displace infected cells as hyperplastic cells migrate to surface
  - Peak 2-3 weeks post-infection
  - Lesions resolve by ~2 months
- Immune response important in clearance/recovery and disease severity
  - CD4, Th1 response, B cells
  - MAIDS, *Veterinary Pathology* 47: 312-317, 2010
    - Don’t clear infection
Citrobacter rodentium

- No known carrier state in immunocompetent
- Recovered mice refractory to infection
- Thick colon, shrunken cecum, no feces
- Long crypts
- Epithelial hyperplasia, colitis cystica
- Loss of goblet cells
- Variable erosion/ulceration and inflammation
Citrobacter rodentium
Citrobacter rodentium
**E. coli**

- Atypical, non-lactose fermenting, gram negative bacilli
- Colonic hyperplasia
  - Segmental
- Immunodeficient mice
- Bacteria in gut lumen, attached to surface and within enterocytes
  - Primary pathogen?
**Helicobacter spp.**

- MAJOR GI pathogens in rodents!
- *H. hepaticus, H. bilis, H. apodemus*
  - Also *H. rodentium, H. muridatum, H. typhlonius, H. ganmani, H. rappini, H. mastomyrinus, H. magdeburgensis, H. pullorum*, etc., etc.
  - Mixed infections
  - Gram negative, spiral bacilli
- Transmission: Fecal-oral, bedding
- Serology
- Fecal PCR preferred (persistent fecal shedding
  - *Helicobacter* genus
  - Differentiate species
**Helicobacter spp.**

- **Pathogenicity**
  - Immunodeficient: Nudes, SCIDS, rag KOs, IL-10 KO
  - A and C3H strains
  - Age (>6mo), sex (M>F), multiple susceptibility genes

- **Proliferative typhlocolitis**
  - Invasive, dysplastic +/- colonic tumors

- **Hepatitis +/- hepatocellular tumors**
  - Biliary canaliculi
  - If kupffer and Ito cell proliferation, think *Helicobacter!

- **Gastritis, gastric atrophy, MALT lymphoma**
  - Also experimental *H. pylori, H. felis, H. suis* infection

- **Rederivation or foster pups (w/in 24 hours) >>> medicated feed**

TC Bracken et al. JAALAS 56: 322-329, 2017

- **Immunodeficient, GEM, C57BL/6**
- ↓ weight gain, embryos/gravid uterus
- ↑ resorptions, intrauterine hemorrhage
Helicobacter spp. – Colitis
Helicobacter spp. – Hepatitis (Early)
Helicobacter spp. – Hepatitis (Late)
**Clostridium piliforme**

- Intracellular, gram negative, spore-forming, filamentous bacilli
- Tyzzer’s disease
- “Species-specific”
- Ingest spores which remain infective for long periods in environment
- Poor husbandry, immunosuppression
- Organisms invade intestinal epithelium → dissemination
**Clostridium piliforme**

- Necrotizing hepatitis
- Necrotizing enterotyphlocolitis
- +/- Necrotizing myocarditis
- Warthin-Starry silver stain
- PCR on feces
- Cortisol provocation test for identification of subclinical carriers
Clostridium piliforme
Salmonella spp.

- *S. enterica* subsp. *enterica*
  - Serovar typhimurium → *S. typhimurium*
  - Serovar enteritidis → *S. enteritidis*
- Rare but common experimental model
- Normal gut microflora = microbial barrier
- Transmission: fecal-oral, fomites +/- vertical
- Fimbrial attachment to M cells → phagocytosis by enterocytes → multiples in GALT and LNs → systemic
  - Readily killed by neutrophils
  - Evade clearance in macrophages
- Liver, spleen, intestines
- Biofilms
- Intermittent fecal shedding → carrier → zoonotic!
Salmonellosis
Salmonellosis
Corynebacterium bovis

- Coryneform-associated hyperkeratosis
- *Immunodeficiency* +/- hairlessness
  - SCID mice are also susceptible!
  - Xenograft studies: ↓ tumor growth; ↑ toxicity of chemotherapeutic agents
- Direct transmission, fomites (*JAALAS* 51: 189-198, 2012)
  - Furred immunocompetent mice, human nasopharynx
  - Airborne dispersal despite individual ventilated caging (IVC)
  - ≥180°F tunnel washer final rinse effective
- Culture or PCR
  - Skin, oropharynx, blood, feces
  - IVC exhaust air duct (EAD) (*JAALAS* 56: 166-172, 2017)
  - SLOW growing – hold for 7 days
Corynebacterium bovis

- Orthokeratotic hyperkeratosis
- Acanthosis
- Sparse dermatitis
- +/- gram positive rods within stratum corneum
- Weight loss
  - Dehydration (trans-epidermal water loss)
  - Anorexia
- DDX: Low ambient humidity; *Staph xylosus* (Vet Pathol 50: 722-726, 2012); cutaneous lymphoma
Corynebacterium + Staphylococcus
**Staphylococcus spp.**

- *S. aureus*
- *S. xylosus*
- Commensal
- Suppurative to pyogranulomatous
  - Splendore-Hoeppli
  - Adnexa (muzzle), periorbital, conjunctiva, preputial glands, LNs
- Necrotizing to ulcerative
  - Burn-like lesions
- Proliferative, hyperkeratotic
- Gram positive cocci
Staphylococcus – Botryomycosis
Staphylococcus – Botryomycosis
Differentials: Trauma, mechanical denudation (cages, waterers), thermal or chemical burns

**Staphylococcus** – Ulcerative Dermatitis
Healing Ulcerative Dermatitis
Differentials: *Corynebacterium kutscheri*, *Streptococcus* spp., *Rodentibacter pneumotropicus* (AKA *Pasteurella pneumotropica*), *Pseudomonas aeruginosa*  
*Exophthalmia!  

**Conjunctivitis, Blepharitis, Retro-orbital Abscesses**
Clitoral/Preputial Gland Abscess

Comparative Anatomy and Histology: A Mouse, Rat and Human Atlas, 2017
Clitoral/Preputial Gland Abscess
Rhinitis/Otitis/Septicemia

- CAR bacillus
- *Bordatella hinzii*
- *Mycoplasma pulmonis*
- *Rodentibacter pneumotropicus* (AKA *Pasteurella pneumotropica*)
- *Pseudomonas aeruginosa*
- *Streptococcus* spp.
- *Burkholderia gladioli, B. cepacia*
- *Klebsiella oxytoca*
Rhinitis/Otitis/Septicemia

- Gram negative bacteria
- Commensals
- Environmental contaminants
- Immunodeficiency
  - C3H/HeJ: defective Toll-like receptor 4
    - Resistant to endotoxins
    - Susceptible to gram negative bacterial infections
Rhinitis
Otitis Media
Viral Diseases
EDIM

- Epizootic diarrhea of infant mice
- EDIM strain of Rotavirus A
- All ages susceptible but disease limited to <2 weeks of age
- Infects terminally differentiated enterocytes in SI/LI
  - Most plentiful and widespread in neonates + slow turnover
  - ↓ #, distribution and differentiation as gut microflora established
- Transmission: fecal-oral; shipping boxes (JAALAS 57: 529-533, 2018)
- Clinical signs in naïve population (lactogenic immunity)
- Complete recovery ~14-17 days of age
- Runting, diarrhea, steatorrhea, pot-bellied
  - Still suckle → visible milk spot!
- Fluid-filled bowel
- Hydropic vacuolar degeneration of enterocytes in villar tips
- Serology, PCR on feces
Vacuolar degeneration of villar tips

Normal vacuolation of neonatal villi
Murine Hepatitis Virus

- Coronavirus
- Multiple strains based on virulence and organotropism
  - Respiratory strains = primary tropism for respiratory mucosa but polytropic (liver, lymphoid tissues); affects immunodeficient
  - Enterotropic strains = selective infection of intestinal epithelium, even in immunodeficient; affects immunocompetent & immunodeficient
- Susceptible strains: C57BL, DBA, nude, SCID, BALB/c
  - Persistent in immunodeficient
- Serology, PCR on feces or mesenteric LN
- Prolonged immunologic effects
- Rederive
Polytropic MHV

- Virulent strains, mice <2 weeks of age, immunodeficient
- CNS, liver, lymphoid tissues, bone marrow, endothelium
  - Necrosis
  - Syncytitia
  - Particularly common in immunodeficient
Polytropic MHV
Enterotropic MHV

- All ages susceptible
  - Disease in neonates
    - Lethal intestinal virus of infant mice (LIVIM)
  - Minimal disease in adult nudes/SCIDs
- All strains susceptible, including those resistant to polytropic MHV
- Necrotizing enteritis
  - Preferential target = ascending colon (?)
- Don’t suckle → dehydrated!
Enterotrophic MHV
Ectromelia Virus

- Mousepox (orthopox)
- Missing limbs
- Pathogenesis
  - Cutaneous invasion $\rightarrow$ local replication $\rightarrow$ regional lymph nodes $\rightarrow$
    primary viremia $\rightarrow$ replication in spleen and liver $\rightarrow$ secondary viremia systemically
- Highly infectious
- Immunocompetent: subclinical; recovery; no carrier state
Ectromelia Virus

- Immunodeficient!
- Acute and rapidly fatal
  - Necrosis and hemorrhage
  - Liver, lymphoid organs, epithelium
- Chronic
  - Cutaneous ulceration and gangrenous necrosis of feet, tail, snout
Ectromelia Virus - Viremia
Ectromelia Virus – Cutaneous
Murine Papillomavirus

- *Veterinary Pathology* 48: 500-505, 2011
- NMRI-Foxn1\textsuperscript{nu} / Foxn1
- PCR $\rightarrow$ novel MusPV
- Transmissible

![Murine Papillomavirus](image)
Differentials: Carcinogens, genotype

Papillomavirus
**Murine Retroviruses**

- Retroelements make up ~37% of mouse genome
  - Mother Nature’s transgenes!
- Endogeneous and exogenous
  - Transmitted via milk, semen, saliva, vertical
  - Most eliminated by rederivation/cross-fostering
- Integrate into genome (proviruses)
  - Random insertional mutagenesis
- Murine Leukemia Virus
  - B or pre-B cell lymphomas/leukemias
  - Granulocytic leukemia
  - Hind limb paralysis/neuronal degeneration in wild strain
  - Hairlessness, premature graying
- Murine Mammary Tumor Virus
  - Bittner agent, MMTV-S, extrachromosomal milk factor = exogenous (C3H)

[Image: http://microbewiki.kenyon.edu/index.php/Koala_Retrovirus]
Mouse Parvovirus

- Minute virus of mice (MVM[p,i,m,c]); mouse parvovirus (MPV1-X) (75%)
- Dependent on S-phase of cell cycle for replication (dividing tissues)
  - Initially replicates in SI intraepithelial lymphocytes, lamina propria and endothelium → systemic
- Immunomodulatory effects on research
- Seroconversion but no clinical disease in immunocompetent
  - Transmission dependent on B cell function vs. T cell function (*JAALAS* 52: 467-474, 2013)
  - Self-limiting – cleared 4 wks post-infection
- Oronasal exposure to urine and feces
  - Contact w/ soiled bedding/cages/food, unsterilized feed
  - Removal of pups from lactating females
- Contaminant of biologicals, patient-derived xenografts
- PCR on mesenteric LNs > feces
  - Hard to eradicate (ventilation systems): rederive/decontaminate
Mouse Kidney Parvovirus

- *Cell* 175: 530-543, 2018
- Inclusion body nephritis
  - Inclusions → tubular degeneration and interstitial fibrosis
  - Cause of death in severely immunodeficient
- Highly divergent parvovirus
- Fecal-oral and urinary-oral transmission
- Widely distributed
Mouse Noroviruses

- Caliciviridae – multiple strains
- Transmission: fecal-oral
- High seroprevalence (35-64%)
- Persistent infection & shedding with no evidence of pathogenicity in immunocompetent
- Replicates in macrophages/dendritic cells & B cells
- Clinical disease +/- mortality in compound immunodeficient mice
  - Acquired (i.e. Rag1KO) & innate (i.e. STATE1KO, IFN αβγR KO) immunity
  - Variable pathology: alveolitis/interstitial pneumonia, hepatitis, splenitis, enteritis, necrosis
  - Impact on research: paneth cell abnormalities, disrupted GI epithelial barrier, increased atherosclerosis, etc.
- Serology, PCR (feces, exhaust plenum)
- Cross foster or rederive
Mouse Astroviruses

- 2nd most common cause of nonbacterial diarrhea in children, elderly and immunocompromised
  - Disrupted intestinal epithelial cell tight junctions → diarrhea
- Dogs, cats, ruminants, pigs, bats, rats
- Mice – wide geographical distribution
  - Immunocompetent and immunodeficient
  - Humoral, innate, cell-mediated immunity
  - Intestines, mesenteric lymph node, spleen, liver, kidney
  - No clinical signs or pathology
  - PCR: Feces, cage swabs, exhaust air debris
- JAALAS 56: 402-411, 2017

Fungal Diseases

http://www.biologyjunction.com/fungi_notes_b1.htm
**Pneumocystis**

- Species specific
  - Mice: *P. murina*
- Ubiquitous
- Asexual: binary fission → trophic form
- Sexual → ascus w/ 8 ascospores
- Pathogenic if immunodeficient
- Asymptomatic or 2° bacterial/viral infections
- Dyspnea, rough hair coat, hunched, cyanosis, death

*PLoS Pathogens 6: e1001009, 2010*
Pneumocystis

- Inhalation of asci
- Ascospores released in alveoli
- Attachment to type I pneumocytes and macrophages by fibronectin-binding integrins
- Necrosis of pneumocytes w/ damage to alveolar basement membranes
- Type II pneumocyte hyperplasia
- Proposed: *Pneumocystis* binds to surfactant protein altering function
Pneumocystis - Immunodeficiency
Parasitic Diseases
Fur Mites = Acariasis

- **Myobia musculi**
  - Head, neck, shoulder
  - Feeds on skin secretions and interstitial fluid
  - Type I hypersensitivity (B6)

- **Myocoptes musculinus**
  - Most common
  - Superficial epidermis
  - Inguinal, ventral abdomen, back

- **Radfordia ensifera** (rat fur mite)

- **Trichoecius rombouti**

- Mixed infections
Fur Mites = Acariasis

- Direct life cycle, ~ 3 weeks long
- Egg, nymph, adult stages on mouse
- Eggs laid on hair shaft
- Direct transmission
  - Suckling mice at 1 week of age
    - Pelage eruption
  - Hairless (nude) mice resistant to experimental infection
Fur Mites = Acariasis

- Alopecia, erythema, pruritus
- Ulcerative dermatitis
- Reduced life span, weight loss, infertility
- Increased total IgE levels (JAALAS 51: 199-208, 2014)
- Skin scrape/tape test
- Pelt exam
- PCR on IVC filter tops
<table>
<thead>
<tr>
<th>MYOBIA</th>
<th>RADFORDIA</th>
<th>MYOCOPTES</th>
</tr>
</thead>
<tbody>
<tr>
<td><img src="MYOBIA.jpg" alt="Image" /></td>
<td><img src="RADFORDIA.jpg" alt="Image" /></td>
<td><img src="MYOCOPTES.jpg" alt="Image" /></td>
</tr>
<tr>
<td>Slightly elongated body</td>
<td>Slightly elongated body</td>
<td>Oval, egg occupies ½ of abdomen</td>
</tr>
<tr>
<td>Bulges between legs</td>
<td>Bulges between legs</td>
<td>Heavily chitinized, pigmented 3rd &amp; 4th legs</td>
</tr>
<tr>
<td>2nd pair of legs: single terminal tarsal claw</td>
<td>2nd pair of legs: 2 terminal tarsal claws of unequal length</td>
<td>Suckers on tarsi</td>
</tr>
</tbody>
</table>
Comparative Pathology and Mouse Phenotyping

**Myobia musculi**
- Bulges between legs
- Slightly elongated body

**Myocoptes musculinus**
- Heavily chitinized pigmented 3rd & 4th tarsi
- 1st pair of legs

♂
♀
Pinworms = Oxyuriasis

- **Syphacia obvelata**
  - Direct life cycle, 12-15 d long
  - Adults in cecum and colon
  - Eggs laid in perianal area; infective within hours
  - Intermittent shedding

- **Aspiculuris tetraptera**
  - Most common
  - Direct life cycle, 23-25 d long
  - Adults in cecum and colon
  - Eggs passed in feces; infective at RT x 6-7 d

- **Syphacia muris** (rat pinworm)
  - Mixed infections
  - Environmental resistance

- Asymptomatic
- Rectal prolapse, intussusception, fecal impaction, diarrhea, poor weight gain, rough hair coat
- Catarrhal enteritis, hepatic granulomas, perianal irritation
- Young, males, immunodeficiency (nude)
- Wild-derived (*Aspiculuris*)
  - JAALAS 56: 42-46, 2017
- Treat > rederive
Pinworms = Oxyuriasis
Pinworms = Oxyuriasis

- PM Gerwin et al. JAALAS 56: 32-41-, 2017
- PCR (fecal pellet & perianal swab)
- Intestinal content exam
- Tape test (Syphacia)
- Fecal float
  - Aspiculuris
    - Bilaterally symmetrical
    - Unembryonated
  - Syphacia
    - Banana-shaped
    - Embryonated
- Swiss roll histology
Demodex musculi

• Outbreaks at 5/63 institutions! (JAALAS 56: 527-533, 2017)
• Commensal → opportunistic in (aged) immunocompromised
  - Impaired Th2 immunity
  - C. bovis hyperkatotic dermatitis & Rodentibacter pneumotropicus retro-orbital abscesses
• Eggs laid in hair follicles → larva → proto/deutonymphs in sebaceous glands → adults (pilosebaceous units)
• Erythema to ulceration, alopecia, blepharitis/conjunctivitis, dermatitis
  - Sebaceous gland hypertrophy → suspect Demodex!
• Interscapular region > dorsum/ventrum > head >> ears
• D. flagellurus: preputial/clitoral gland via sexual/direct contact
Demodex musculi

- Deep skin scrapes + fur plucks (eggs/larva), skin histology
  - 2 sites
  - PCR on pelt swabs, IVC (Idexx Bioresearch)
  - Skin fragmentation digestion for ID of life stages

- Treatment
  - Amitraz baths (biweekly) $\rightarrow$ ↓ mite burden but no effect on clinical signs
  - Oral ivermectin (feed) $\rightarrow$ ↓ mite burden
  - Topical moxidectin/imidacloprid (q wk x 8)
  - Rederive

Demodex musculi

JAALAS 57: 173-185, 2018; guanine concretions
**Spironucleus muris**

- Flagellated binucleate protozoan w/ direct life cycle
- SI, cecum
- Commensal
- Diarrhea in young, stressed, immunodeficient
- Trophozoites ± crypt dilation, crypt abscesses, lymphoplasmacytic inflammation
- Recovery → asymptomatic carriers
- Interspecies (hamster) transmission

http://dora.missouri.edu/mouse/spironucleus-muris/
Tapeworms

- Rare
- *Rodentolepis nana*
- *Hymenolepis diminuta*
- *Rodentolepis microstoma*
- Zoonotic, also direct lifecycle
- Arthropod (beetles, fleas, moths) intermediate host
- Cysticerci in lamina propria, adults in lumen

Pet Store Mice

- **JAALAS 51: 37-41, 2012**
- MHV 18/18
- MPV 1/2 – 14/18
- PVM – 2/18
- MVM – 10/18
- EDIM – 8/18
- *C. piliforme* – 3/18
- Fur mites – 11/18; dual infections
- Pinworms – 7/18
- *Rodentolepsis nana* – 9/18
- *Rodentibacter pneumotropicus* (AKA *Pasteurella multocida*) – 5/18
- MDR B-hemolytic *Enterococcus faecium* – 3/18
- Peripheral eosinophilia – 6/18

[http://www.vintagelpscollector.com/Collections/petpresents.htm](http://www.vintagelpscollector.com/Collections/petpresents.htm)
Mouse Neoplasia

- Strain and substrain
- Age
- Environment
  - Stress
  - Temperature
  - Bedding
  - Cage density
  - Altitude
- Diet
- Reproduction
  - Parity
  - Lactation
- Endogenous retroviruses
- Concurrent disease
- Experimental manipulation
Mouse Neoplasia

- Genetically engineered mice (GEM)
  - Mouse Models of Human Cancers Consortium (MMHCC)
    - Consensus report & recommendations
    - Lymphoid, nonlymphoid, mammary, prostate, lung, CNS, PNS, nerve sheath, intestine, exocrine pancreas
  - Histology may be different from neoplasms arising spontaneously
    - Signature phenotypes
      - ‘Pathway pathology’
      - Wnt vs. ErbB2 vs. Myc

http://jewelsonhampton.com/loose-diamonds-rubies-emeralds-sapphires/
Mouse Neoplasia

• Intraepithelial neoplasia
  - Atypical epithelial cells, dysplasia
  - Precursor to cancer (molecular changes)
  - May disappear, remain unchanged, progress to cancer (hi grade)
  - AIN, GIN, BilIN, MIN, PIN, PanIN, EIN, CIN, VaIN, VIN, PeIN

• Cellular transitions
  - Epithelial to mesenchymal transition
  - Mesenchymal to epithelial transition
  - Endothelial to mesenchymal transition

Carcinogenesis 27: 1054-1067, 2006
Lymphomas & Leukemias

- B cell: Small, splenic marginal zone, follicular, diffuse large, histiocyte-associated DLBC, lymphoblastic, plasmacytoma
- T cell: Precursor lymphoblastic, small
- 129, AKR, CFW, C58, CB17.scid, NOD/scid, BALB/c
- Spontaneous, MuLVs, chemicals, irradiation, genetically engineered
- Leukemia: Blood, bone marrow vs. spill-over
- Acute tumor lysis syndrome
Lymphomas & Leukemias
Plasmacytomomas

- Intraperitoneal injection of mineral oil, pristane, plastics, other foreign material
- BALB/c mice
Granulocytic Leukemia

- Retroviruses, carcinogens, irradiation
- Spleen → widespread
- Anemia
- WBC >200,000
- Grossly may appear green
Acute Tumor Lysis Syndrome

- Rapid destruction of malignant cells → massive release of cellular contents/breakdown products → acute metabolic crisis
  - High P, K, uric acid
  - Hypocalcemia
- Death → cardiac arrest, hypoxia
- Lymphoid neoplasms, granulocytic leukemia
- Chemotherapy, corticosteroids, hormones, cytokines
- Spontaneous (↑ tumor burden)
Histiocytic Sarcomas

- SJL and aging B6
- Reticulum cell sarcoma, histiocytic lymphoma, MFH, malignant histiocytosis, Kupffer cell sarcoma
- Incidence: 1-22% depending on strain
- Liver, uterus
- Mononuclear-phagocytic cells
  - MNGC, erythrophagocytosis
- Hyaline droplets → kidney
  - Overproduction of lysozyme → saturation of lysosomal hydrolytic enzymes
  - Granulocytic leukemia, B cell LSA
- EMH → spleen, liver
- Dysmyelopoiesis (↓M; ↑E) → BM
Hepatocellular Lesions

- Foci of cellular alteration (basophilic, eosinophilic, clear)
  - Altered staining properties, no alterations in liver architecture, no compression of parenchyma
- Regenerative hyperplasia
  - Prior or ongoing hepatocellular damage
- Hepatocellular adenoma/hepatoma and carcinoma
  - Trabecular, solid, adenoid
- Hepatoblastoma
- Cholangiomas, cholangiocarcinomas, mixed (rare)
- Spontaneous (A and DBA mice, rats), hepatocarcinogens, *Helicobacter hepaticus*
Foci of Cellular Alteration

- Phenobarbital: ↑ γ-glutamyl transferase
- Diethylnitrosamine: ↓ glucose-6-phosphatase

International Classification of Rodent Tumors
Hepatocellular Tumors

Adenomas

Carcinomas
Hepatocellular Tumors

Nodular Hyperplasia

Adenoma

Carcinoma
Hepatoblastomas

- Rare (<1%)
- Arise within or adjacent to adenomas/CAs
- Distant mets, including lungs
- Diethylnitrosamine / phenobarbital
- Cell of origin?
  - Oval, hepatocytes, biliary epithelium
  - Rosettes
Pulmonary Tumors

- Common in older mice
- A, BALB, FVB, 129, B6;129 strains
  - K-ras
  - Viral infections, chemicals, age
- Bronchogenic tumors → hilus
  - Not reported in mice
- Bronchiolar/alveolar tumors → periphery
  - Bronchiolar tumors originate from Club (Clara) cells
  - Alveolar tumors originate from type II pneumocytes → surfactant production
  - Current recommendation: pulmonary adenoma or carcinoma, specify solid, papillary or mixed
  - Hyperplasia → adenomas > carcinomas
  - Must differentiate from metastatic tumors!
  - Concurrent acidophilic macrophage pneumonia
Mammary Gland Tumors

- Strain dependent → C3H, A, DBA, BALB/c
- Murine Mammary Tumor Viruses (retrovirus)
  - Endogenous
  - Exogenous – removed by rederivation
  - Bittner agent – vertical transmission (milk)
- Carcinogens, prolactin, estrogen, progesterone
- Stress, *ad libitum* feeding, multiparity
- Glandular, acinar, cribriform, papillary, solid, squamous, fibroadenoma, NOS, adenomyoepithelioma, adenosquamous
Mammary Gland Tumors
FVB/N Mice

- Persistent mammary gland hyperplasia +/- squamous nodules
- Hyperplasia or adenoma of pars distalis
  - Acidophils/lactotrophs
- Upwards of 50% incidence by 18 mo.
- Mammary carcinomas in multiparous mice with pituitary prolactinomas
  - *Veterinary Pathology* 46: 736-745, 2009
  - Adenosquamous differentiation
  - Epithelial to mesenchymal transition
  - ERα immunoreactivity
FVB/N Mammary Hyperplasia/Squamous Nodules

(Ignore the color of this mouse!)
Pituitary Gland Tumors

- FVB/N: 50% incidence by 18 months
- Persistent
- Hyperplasia or adenoma of pars distalis
  - Acidophils/lactotrophs
- Mammary carcinomas in multiparous mice with prolactinomas
  - Adenosquamous differentiation
  - EMT
  - ERα immunoreactivity

Comparative Medicine 53: 424-432, 2003
Pituitary Gland Tumors

- Hyperplasia
- Adenoma
- Carcinoma
Salivary Gland Myoepitheliomas

- Multiple strains of mice, especially BALB
  - 8/24 nonthymic tumors in SCID mice
    (Comp Med 61: 227-234, 2011)
- Females > males
- Submaxillary and parotid
- +/- mammary, preputial and Harderian gland tumors
- Pulmonary metastasis with large tumors
- Myeloid hyperplasia → tumor secretory product
Harderian Gland Tumors

- Hyperplasia $\rightarrow$ adenoma $\rightarrow$ adenocarcinoma
  - Papillary, cystic, acinar
- Unilateral or bilateral
- Exophthalmia
- Old albino and pigmented strains
  - 0.5-14.9% incidence
- Spontaneous, irradiation, carcinogens
- Mets to lungs
Harderian Gland Tumors

Adenoma

Adenocarcinoma w/ lung mets
Hemangiomas/Hemangiosarcomas

- Anywhere!
- Liver, uterus, skin, spleen, bone marrow
- Ovary
  - DDX: Angiectasia
  - Thrombosis seen in both
- Multicentric or metastasis?
- Metastasis to lungs
Angiosarcomas
Osteomas/Osteosarcomas

- Osteoma
  - Periosteum
  - Skull
  - F > M
  - Multicentric in outbred OF-1, CF-1
- Osteosarcoma
  - Metaphysis
  - Eburnating, osteoblastic, fibroblastic, osteoclastic, chondroblastic, vascular, anaplastic, mixed
  - Metastasis to lung, liver, spleen
    - Especially caudal vertebral
  - NOD mice
Osteosarcomas
SQ Neural Crest Tumors (FVB Mice)
Teratomas (esp. 129 Mice)
Teratomas (129 Mice)

Neuro/Ectoderm

Mesoderm

Endoderm
Differentials?

- *Helicobacter* spp.
- *Citrobacter rodentium*
- Atypical *E. coli* in immunodeficient mice
- Pinworms
  - *Syphacia obvelata*
  - *S. muris*
  - *Aspiculuris tetraptera*
Differentials?

- Rotavirus A
- Enterotropic mouse hepatitis virus
- *Salmonella* spp.
- *Clostridium piliforme*
- Reovirus-3
- Mouse Norovirus (immunodeficient)
- *Spironucleus muris*
Differentials?

- Circling, rolling
- Necrotizing polyarteritis
- Otitis media
- Brainstem infarction (Swiss Webster)
- Brain neoplasm
- Genotype

*Neuroscience Research 72: 296-305, 2012*
Differentials?

- Fur mites
  - *Myobia musculi*
  - *Myocoptes musculinus*
  - *Radfordia affinis*
  - *R. ensifera* (rat fur mite)
  - *Trichoecius rombouti*
- *Staph* or *Strep* spp.
- Trauma/fighting
- Ulcerative dermatitis of *B6 mice*
‘Do It Yourself Pathology’

<table>
<thead>
<tr>
<th>Phenotypes</th>
<th>mPer2&lt;sup&gt;mm&lt;/sup&gt; Mice (18 months old) (n = 20)</th>
<th>Wild-Type Mice (18 months old) (n = 20)</th>
<th>mPer2&lt;sup&gt;mm&lt;/sup&gt; Mice 16 months after IR (n = 14)</th>
<th>Wild-Type Mice 16 months after IR (n = 20)</th>
<th>p value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salivary gland hyperplasia</td>
<td>20 (50%)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0</td>
<td>14 (100%)</td>
<td>1 (5%)</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>Teratoma in male mice</td>
<td>10 (100%)</td>
<td>0</td>
<td>9 (100%)</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Hair graying 6 months after IR</td>
<td>3 (15%)</td>
<td>0</td>
<td>10 (71%)</td>
<td>0</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>Lymphoma</td>
<td>0</td>
<td>0</td>
<td>2 (10%)</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

*Six irradiated mPer2<sup>mm</sup> mice were lost at 9 months after irradiation during the summer flooding in Houston in 2001.

<sup>a</sup>50% of mPer2<sup>mm</sup> mice showed enlarged salivary glands by physical examination. At autopsy, all the mPer2<sup>mm</sup> mice older than 8 months of age were found to have salivary gland hyperplasia.
Teratomas

Neuro/Ectoderm  Mesoderm  Endoderm
Preputial/Clitoral Glands

Comparative Anatomy and Histology: A Mouse, Rat and Human Atlas, 2017
‘Do It Yourself Pathology’

The Circadian Gene *Period2* Plays an Important Role in Tumor Suppression and DNA Damage Response In Vivo

Erratum

The Circadian Gene *Period2* Plays an Important Role in Tumor Suppression and DNA Damage Response In Vivo

In the article by Fu et al. (Cell 111, pp. 41-50), Figures 1c and 1d show abnormally enlarged preputial glands with severe duct ectasia, focal hyperplasia, and hyperkeratosis, rather than teratoma. This revised diagnosis also applies to Table 1 on page 43. The conclusion of the study remains unaffected by this correction.

We thank Dr. J.M. Ward for his notification and the revised diagnosis.

Loning Fu, Helene Pellicano, Jinsong Liu, Peng Huang, and Cheng Chi Lee

It’s NOT a Tumor!!!!
Volume III = Systems Toxicologic Pathology
References – 2
Comparative Pathology and Mouse Phenotyping

References - 3
References - 4

- National Toxicology Program Nonneoplastic Lesion Atlas
- International Harmonization of Nomenclature and Diagnostic Criteria,
  [www.toxpath.org/inhand.asp](http://www.toxpath.org/inhand.asp)
- Proliferative & non-proliferative lesions of mice & rats:
  - Cardiovascular
  - Bone, joints, teeth
  - GI tract, pancreas, salivary glands
  - Female reproductive system
  - Male reproductive system
  - Soft tissue, skeletal muscle, mesothelium
  - Integument
  - Mammary, Zymbal's, preputial, clitoral glands
  - Urinary system
  - Central and peripheral nervous systems
  - Hepatobiliary system
  - Respiratory tract
  - Endocrine
  - Special Senses
References - 5

• MMHCC Consensus/recommendation papers for proliferative lesions in GEMMs
  ✓ LD Berman-Booty et al, *Toxicologic Pathology* 40: 5-17, 2012
Acknowledgements

- Sheree Beam
- Charlie Clifford
- Chelsea Martin
- Hai Nguyen
- Brett Saladino
- Trenton Schoeb
- Teresa Southard
- Uncited Individuals
- Cornell University College of Veterinary Medicine/Dr. John M. King’s Necropsy Show & Tell
- U. of Missouri/IDEXX Bioresearch

- Michael Eckhaus
- Duncan Russell
- Nozomi Shimonohara
- Paul Stromberg
- Jerry Ward
- ACLAM Lab Animal Medicine and Science Series II
- Joint Pathology Conference - VSPO
- Noah’s Arkive
- The Jackson Laboratory
- Dean Percy – Pathology of Laboratory Rodents and Rabbits, 4th Edition
Questions?

Laperle.1@osu.edu
www.vet.osu.edu/CPMPSR