Animal Research Biosafety
Introduction

Animal Research at MIT
Specific practices and procedures to protect the animals, researchers & caretakers
Possible risks and exposures when working with animals
Rules for working with animals in DCM Facilities and Laboratories
Animal Research Biosafety
MIT Animal Research

- There are at least 8 animal facilities in 5 buildings
- All are centrally managed by DCM [Division of Comparative Medicine]
- Program is AAALAC Accredited, Exemplary
AAALAC Accreditation

Association for Assessment and Accreditation of Laboratory Animal Care International
AAALAC Accredited Program

- Committee on Animal Care: protocols, information & review & approval, policies
- DCM: provision of appropriate animal care, features of facility, animal handling & surgery training, veterinary care
- EHS Office: training and risk assessment (IHP, RPP, BSP)
- Researchers: compliance with all policies, rules, training
- CAB/ESCRO policies, RPP policies
- Associate Provost/VP for Research is Institutional Official
Rules for Animal Care and Use

- The need for use of animals must be clear and unequivocal.
- The minimum number of animals must be used, consistent with experimental needs, and no alternative exists.
- The animal model must be appropriate to the experiment.
- Pain must always be minimized: treat for pain prior to and after surgeries.
- Must use sterile technique and instruments in a designated area.
- Must adhere to DCM standards for surgery, recovery, and completed all trainings.
- Animals must not undergo undue stress, all stress inducing procedures must be specially reviewed and approved by CAC.
Rules for Animal Care and Use

- Animals must be housed in facilities able to maintain appropriate temperature and humidity
- Kept clean, watered and well fed, if injured all must be supplied in an easily reached manner
- A strict health assurance program must be in place: sentinel animals as well
- The facility must be designed to prevent cross contamination of air from room to room and from public areas into animal facilities
- Must be kept clean and free of sources of infection
Animal Research Facilities and Possible Risks
Animal Room
Potential exposures in animal facilities:

- **Allergies:** exposure to animal dander
- **Infections:** Bacteria, viruses, or chemicals used in animal studies
- **Physical injury:** Noise, slips, trips, animal bites, scrapes, scratches
- **People are a health risk to animals:** e.g. TB, flu, measles, stomach diseases
Types of animals and their specific risks:

- Mice, rats, rabbits, ferrets: bites, allergies, scratches
- Birds: scratches, Salmonella, bites
- Amphibians: Salmonella infection, bites
- Sheep & pigs: Q fever infection, physical injury
- Primates: scratches, bites, and infections: e.g. Salmonella, Campylobacter, Shigella, Yersinia, TB, Simian B virus
To Be Exposed

- You would have to:
  - Touch the animal or the dirty bedding with bare hands and transfer to face, eyes or cut
  - Handle or work with animals directly
  - Get cuts, scrapes, splashes, splatter from animals or equipment used on animals
  - Breath dusty air (without mask)
  - If you wear the required PPE and wash your hands after removing PPE: low low RISK
Exposures are minimized:

- Animals have thorough initial health screenings and regular monitoring for diseases (see next slides about primates)
- Following Standard Practices and DCM procedures including use of clearly described PPE
- Controls on access to animal areas
- Strict control and containment procedures for special animal experiments (viruses, bacteria, chemicals, radiation)
- Thorough cleaning procedures for animal equipment, cages
- Strong training program and oversight for DCM staff
- Health screening: occ health requirments, DCM staff and researchers that works in a primate room must have annual tuberculosis screening and vaccinations such as tetanus
Example: non-human primates [NHP]

- NHP are screened for diseases before being allowed to come here
- Complete physical is done & each animal’s medical file accompanies it
NHP surveillance: after arriving at MIT

- All animals quarantined 10 weeks, more tests done before release for use
- Extensive and repeated testing done: physical exam, weight and body condition, blood count, blood chemistry, SIV, simian B virus, STLV-1, TB, Measles, SRV, Salmonella, Shigella, Campylobacter, parasites, chest radiograms, skin diseases
Designated Areas

• Special Areas: look for signs
  − Non-human primate rooms
  − Special mice area (animals are specially sensitive)
  − ABL 2 locations: bacteria/viruses and chemicals given to animals

• These Special Areas have special procedures:
  − Special disinfection procedures for cages, equipment
  − Special PPE
  − Special cages or racks

• Dress Like What’s Posted on the Doors
Designated Areas

• Before entry to Special Areas:
  − change into a different gown, gloves, shoe and hair covers, masks, eye and face shields (possibly) depending on animals and room use

• After leaving Special Areas:
  − Once done or leaving room or area, take off special PPE & dispose upon leaving special area, & put back on standard PPE, WASH HANDS
  − Decontaminate equipment before leaving special areas
Quarantine Facility

- HAIR COVER
- DISPOSABLE COAT
- GLOVES
- SHOE COVERS
Non-human Primate Room

- HAIR COVER
- FACE MASK
- Safety Glasses and Face Shield
- DISPOSABLE COAT for that room only, often a different color
- Gloves (2)
- Shoe Covers
HAIR COVER

DISPOSABLE LAB COAT

FACE MASK

GLOVES

SHOE COVERS

Special Rooms, note mask
Animal Protocol and Biosafety Review Process
Role of CAB/ESCRO

- Policies for safe use of biological materials
- Policies developed to cover animal research containment
- Integration with other Institutional committees (IACUC, IRB)
- CAB/ESCRO is Early Warning System
CAB: Bench to Bedside Involvement

<table>
<thead>
<tr>
<th>Institute Committee</th>
<th>Isolation of gene</th>
<th>In vitro studies, pathogen</th>
<th>In vivo testing animal</th>
<th>Animal model of disease</th>
<th>Preclinical studies</th>
<th>Clinical Trial</th>
</tr>
</thead>
<tbody>
<tr>
<td>CAB</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
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<td>+</td>
</tr>
<tr>
<td>IACUC</td>
<td></td>
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<td>+</td>
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<tr>
<td>COUHES</td>
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</table>
Animal Research

- Mice, rats
- Rabbits, ferrets, frogs
- Sheep, pigs
- Monkeys

- Genetic diseases, cancer, disease models
- Surgery, disease models, vision studies
- Cardiovascular & heart disease
- Learning & memory, brain function
Types of Animal Research

• Vaccine development
• Understanding the Immune system
• Pathogen virulence factors and disease
• Arterial structure and coronary disease
• Brain development and neural structure
• Injury & joint treatments
Animal Research

- Mode of injection, ip, sc, iv, brain, etc.
- Data on vector prep characterization including use in animals at least once.
- Need for high containment after vector clearance from animal is case by case.
- DCM staff are able to handle transgenics after certain time.
Animal Research Biosafety

- Risk to investigators
- Risk to animal handlers
- Risk to other animals
- Shedding, exhalation (mode of transmission)
- Duration of expt and shedding
What Can We Control?

- Type of room: containment level
- Type of cage
- Cleaning & handling procedures, PPE
- Bedding and cage cleaning
- Tissue harvest
- Disposal
- Training: DCM and researchers
Pathogens in Animals

Infects humans & is shed
- ABL2, IVC, administration in BSC, PPE
- Researchers take care of animals for duration
- Bedding incinerated, cages chemically sterilized before washing
- Occ Health

Infects animals & is shed
- ABL2, IVC, administration in BSC, PPE
- Researchers take care of animals for duration
- Bedding incinerated, cages chemically sterilized before washing
- Occ Health
<table>
<thead>
<tr>
<th>Gene transfer vector</th>
<th>Host range&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Insert or gene function&lt;sup&gt;c&lt;/sup&gt;</th>
<th>Laboratory Biosafety Containment Level&lt;sup&gt;d&lt;/sup&gt;</th>
<th>Animal Biosafety Containment Level&lt;sup&gt;e&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>MMLV based - gag, pol, env deleted</td>
<td>Ecotropic</td>
<td>S, E, M, G, CC, T, MP, DR, R, TX, O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;</td>
<td>BSL 1*</td>
<td>ABSL1, infected or transgenic animals housed separately</td>
</tr>
<tr>
<td></td>
<td>Amphotropic, VSV-G pseudotyped</td>
<td>S, E, M, MP, DR, T, G, O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;, R, CC</td>
<td>BSL 2</td>
<td>Infection at BSL2, for embryo injection for transgenic or knockouts; all infected animals held at ABSL2 for 5 days, cage change at +5 days, then animals can be held at ABSL1 for duration of experiment</td>
</tr>
<tr>
<td>Herpes virus based: non-lytic replication defective</td>
<td>Strong cell type specificity but a broad host range across species (vertebrates)</td>
<td>S, E, M, MP, DR, T, G, O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;, R, CC, TX</td>
<td>BSL 2</td>
<td>Infections done at BSL2, infected animals remain at ABSL2 for duration of experiment.</td>
</tr>
<tr>
<td>Lentiviral based – HIV, SIV, EIAV, FIV, etc.; gag, pol, env, nef, vpr deleted, addn safety features such SIN LTR deletions</td>
<td>Ecotropic, amphotropic: VSV-G pseudotyped, infects many cell types and wide host range</td>
<td>S, E, M, MP, DR, T, G, O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;, R, CC, TX</td>
<td>BSL 2 or 3</td>
<td>No toxin in vector</td>
</tr>
<tr>
<td>Adenovirus based; serotype 2, 5, 7; E1A &amp; B deleted, along with E3 or E4 deleted in some systems</td>
<td>Broad host range, infective for many cell types</td>
<td>S, E, M, T, MP, DR, R, G, CC</td>
<td>BSL 2</td>
<td>Injections done at BSL2, after +5 days cage change injected animals may be held at ABSL1</td>
</tr>
<tr>
<td>Alphavirus based -- SFV, SIN</td>
<td>Broad host range</td>
<td>S, E, M, T, MP, DR, R, G, CC</td>
<td>BSL 2</td>
<td>Persistent variants, use of stable PCLs, no RCV detected, inject at BSL2, then ABSL1</td>
</tr>
<tr>
<td>Baculovirus based</td>
<td>Broad mammalian host cell range depending on packaging</td>
<td>S, E, M, T, MP, DR, R, G, CC</td>
<td>BSL 1*</td>
<td>BS1 for injection, ABS1 for animals</td>
</tr>
<tr>
<td>AAV based - rep, cap defective</td>
<td>Broad host range, infective for many cell types including neurons</td>
<td>S, E, M, T, MP, DR, G, O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;, R, CC</td>
<td>BSL 1*</td>
<td>Preps must be clear of helper virus, ABS1 after vector clears from animal</td>
</tr>
<tr>
<td>Poxvirus based-Canarypox, Vaccinia virus</td>
<td>Broad host range</td>
<td>S, E, M, T, DR, MP, CC, R, G</td>
<td>BSL 2</td>
<td>BS2 for injection: depending on virus and transgene, injected mice may be ABS1 or 2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>O&lt;sub&gt;v&lt;/sub&gt;, O&lt;sub&gt;c&lt;/sub&gt;,</td>
<td>BSL 2+</td>
<td>BS2 for injection: depending on virus and transgene, injected mice may be ABS1 or 2</td>
</tr>
</tbody>
</table>

<sup>a</sup>Gene transfer vector

<sup>b</sup>Host range

<sup>c</sup>Insert or gene function

<sup>d</sup>Laboratory Biosafety Containment Level

<sup>e</sup>Animal Biosafety Containment Level
1. Standard Policies
2. Work starts at the laboratory level to establish acceptable parameters for materials to be used in animals
3. Animal researchers follow sets of standard practices
4. Constant review of acceptable parameters
5. Safety & Responsible Conduct of Research
### Characteristics of viral systems commonly used for recombinant gene transfer

<table>
<thead>
<tr>
<th>Virus Type</th>
<th>Nucleic Acid</th>
<th>Envelope d</th>
<th>Route of Transmission</th>
<th>Genome Size</th>
<th>Insert Size</th>
<th>Clinical Use</th>
<th>Risk Group</th>
<th>Integration</th>
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</thead>
<tbody>
<tr>
<td>Poxvirus</td>
<td>dsDNA</td>
<td>Y</td>
<td>Aerosol, Direct Contact</td>
<td>192kb</td>
<td>&gt;90Kb</td>
<td>Y</td>
<td>2</td>
<td>N</td>
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<tr>
<td>Retrovirus</td>
<td>diploid ssRNA</td>
<td>Y</td>
<td>Blood-borne</td>
<td>8.3kb</td>
<td>7Kb</td>
<td>Y</td>
<td>1/2</td>
<td>Y</td>
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<tr>
<td>Lentivirus</td>
<td>diploid ssRNA</td>
<td>Y</td>
<td>Blood-borne</td>
<td>9.7kb</td>
<td>8Kb</td>
<td>N</td>
<td>3</td>
<td>Y</td>
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<tr>
<td>Adenovirus</td>
<td>dsDNA</td>
<td>N</td>
<td>Aerosol</td>
<td>36kb</td>
<td>8-30kb</td>
<td>Y</td>
<td>2</td>
<td>N</td>
</tr>
<tr>
<td>Adeno-Associated Virus</td>
<td>ssDNA</td>
<td>N</td>
<td>Aerosol</td>
<td>4.5kb</td>
<td>4Kb</td>
<td>Y</td>
<td>1</td>
<td>Y/N°</td>
</tr>
<tr>
<td>Alphavirus</td>
<td>ssRNA</td>
<td>Y</td>
<td>Blood-borne</td>
<td>11.7kb</td>
<td>7-8Kb</td>
<td>Y</td>
<td>2/3</td>
<td>N</td>
</tr>
<tr>
<td>Baculovirus</td>
<td>dsDNA</td>
<td>Y</td>
<td>Direct contact</td>
<td>90-160kb</td>
<td>large</td>
<td>N</td>
<td>1</td>
<td>Y</td>
</tr>
<tr>
<td>Herpesviruses</td>
<td>dsDNA</td>
<td>Y</td>
<td>Direct contact</td>
<td>152kb</td>
<td>100Kb</td>
<td>Y</td>
<td>2</td>
<td>N</td>
</tr>
</tbody>
</table>
Typical Animal Care Facility

Arrows indicate work flow direction
Simian B virus: information

- Non-human primates (macaques) can present risks to people e.g. latent simian B virus
- Rare disease, and very rare cause of researcher illness
- Important to note: All MIT macaques are repeatedly screened and none have been found to secrete this virus
Simian B Virus: route and sources of exposure

- Bite, scratch from primate directly
- **Also:** splatter, splash to eyes, nose or mouth, etc.
- Cuts or scratches from contaminated equipment, cages, and needlesticks
- The following can cause infection: tissues, cells, saliva, feces from infected animals, but must have contact with your broken skin, eye, nose, or mouth
Designated Areas

• These facilities have high ventilation rate, and ABL-2 cages prevent release of bacteria or viruses and chemicals outside of cages

• Bacteria/viruses and chemicals rigorously controlled during use and in all animal handling procedures and waste disposal