

# Wet Lab for Mice

- **Requirements for Working with Animals**
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- **Biology of the Mouse**
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*While working with your animals, it is important to be aware of the heavily regulated animal research environment and threats. There are federal regulations, state regulations, and UCLA- ARC (IACUC) policies. There is also pressure from the outside world and the animal rights movement. It is important to strive to employ the 3R's - Replacement, Reduction and Refinement.*

*DLAM is here to ensure proper care and treatment of your animals. This class will explain the biology of mice, the health issues that arise, and the health reporting system used by DLAM. The lab should be able to recognize and manage unhealthy animals. It is the responsibility of the lab to respond to all animal health issues. The lab should also be cognizant of the mistreatment of animals and know how this is handled at UCLA and by the ARC.*

# Training Requirements for Working with Animals at UCLA

Revised 5/13/13

## 1. ARC General Certification ([CITI](https://www.citiprogram.org/) Program Training)

<https://www.citiprogram.org/>

All personnel listed on an ARC protocol (including all Principal Investigators and Faculty Sponsors, even if they will not have direct animal contact) must complete the online CITI "Animal Research" training course prior to approval to work with animals. In addition, all active animal users must be re-certified every three years.

**NEW** animal users must complete the [CITI](#) training prior to being approved to work with animals, and will need to be re-certified every three years thereafter.

As of September 1, 2009, all **CURRENT** animal users must be re-certified three years after the date of their previous certification. (For example, if you completed certification on July 1, 2008, your certification to work with animals will expire on July 1, 2011.)

***Please note that, as of September 1, 2009, prior exemptions to the ARC Certification Test are no longer honored.*** However, certifications and exemptions for species-specific training and aseptic surgery will remain valid. For more information: <http://oaro.research.ucla.edu/certification-info>

## 2. Medical History Questionnaire (MHQ):

<http://mhq.healthsciences.ucla.edu/>

All UCLA staff and/or students who handle animals or animal tissue, or who have access to the vivarium, must complete this form each year. All questions are required.

## 3. Mouse Users Training:

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

All mouse users must complete two phases of training:

1. Complete the online course entitled "Working with Mice in Research Settings", which is available via the DLAM website (see link above).
2. Once the online course has been completed, you will be able to print your certificate of completion and enroll in the next available session of the hands-on portion: Mice Users Wet Lab.

## 4. Rat Users Training:

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

All rat users must complete two phases of training:

1. Complete the online course entitled "Working with Rats in Research Settings", which is available via the DLAM website (see link above).
2. Once the online course has been completed, you will be able to print your certificate of completion and enroll in the next available session of the hands-on portion: Rat Users Wet Lab.

## 5. Barrier Training (required for all mouse and rat users):

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

All rodent users that will require barrier facility access will typically require two phases of training:

1. Complete the online course entitled "Working in a Barrier Facility", which is available via the DLAM website (see link above). ***Note: A certificate from this online course is also a prerequisite for other barrier walkthroughs (CHS 6V Rat, Warren Hall and TLSB).***
2. Once the online course has been completed, you will be able to print your certificate of completion and enroll in the next available session of the CHS B-Floor Barrier Walkthrough.

***For areas that do not currently have online enrollment, contact the area supervisor for a guided tour.*** The Division of Lab Animal Medicine (DLAM) maintains the following areas:

### **Lab Access Barriers:**

NRB-3/4/ floor (B09 suite)  
Gonda  
MRL  
Warren Hall  
JSEI  
CHS / PHB-B Floor  
2V CHS  
6V CHS  
BRI  
TLSB

### **DLAM Maintained Barriers (no lab access):**

BSRB - Arch (suites)  
4<sup>th</sup> Floor- CHS  
NRB-B Floor (B05 suite)  
5 rooms-CHS B Floor (BV366, 518, 613, 623, 625, service 1/2 B6:065)  
Tier 3  
Rehab  
ART Lab

### **Facilities Run by DLAM:**

Franz Hall  
Life sciences  
Slichter Hall  
Rehab

### **Biocontainment Facilities:**

CNSI  
NRB 3<sup>rd</sup> floor (2 rooms)  
3V CHS  
BSRB (specific area)  
AR-173

## 6. Aseptic Surgical Techniques (required if performing or assisting with survival surgery).

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

Two phases of training are required:

1. Complete the online course entitled "Aseptic Surgical Techniques", which is available via the DLAM website (see link above).
2. Once the online course has been completed, you will be able to print your certificate of completion and enroll in the next available session of the hands-on portion: Aseptic Surgical Techniques (Wet Lab).

**7. Suture Training for Beginners** (optional training *only* for those that will be performing or assisting with surgery).

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

Two phases of training are required:

1. Complete the online course entitled "Suture Training for Beginners", which is available via the DLAM website (see link above).
2. Once the online course has been completed, you will be able to print your certificate of completion and enroll in the next available session of the hands-on portion: Suture Training for Beginners (Wet Lab).

***Note: both the online and hands-on portions of Suture Training for Beginners must be complete in order to take the Advanced Suture Training Class (currently offered by appointment only).***

**8. Biocontainment Training (required for access to all biocontainment facilities):**

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/default.aspx>

Three phases of training are required:

1. Complete the online course entitled "Working in a Biocontainment Facility", which is available via the DLAM website (see link above).
2. Complete the required EH&S Classes:  
Biosafety A, B, Cs - Biosafety Level 2 (BSL2)  
Biosafety Level 2 with Biosafety Level 3 practices (BSL2+)  
Biological Safety Cabinet (BSC)  
Bloodborne Pathogens (BBP)  
Medical Waste Management (MWM)

*Please contact EH&S to obtain a copy of your current transcript:*

[training@ehs.ucla.edu](mailto:training@ehs.ucla.edu)

3. Print your certificate of completion for the online DLAM course (step 1) and enroll in the next available session of the CHS 3V Biocontainment Barrier Walkthrough. *Note: the CNSI BSL2 Barrier Walkthrough requires a separate verification of requirements before enrollment is allowed.*

**Note: You must bring the certificate of completion for the online course "Working in a Biocontainment Facility" and proof of completion (e.g. transcript) for all required EH&S courses. Walkthroughs will not be carried out if the requirements listed above are not met. Failure to provide proof of EH&S training will result in subsequent re-enrollment block and/or a \$50 recharge to your PI's account.**

**8. Species Specific Training/ Procedure Specific Training:**

Contact DLAM Training: [dlamtraining@mednet.ucla.edu](mailto:dlamtraining@mednet.ucla.edu)

**9. Your name must be listed on an active protocol:**

An amendment to the protocol must be submitted to the ARC by the Principal Investigator. <http://oaro.research.ucla.edu/calendar>



**10. Vivarium Access:**

Go to the Bruin card website to set up an account and a pin number:

<http://secure.bruincard.ucla.edu/bcw/web/Home.aspx>

(Access is granted once you are on the active protocol.)

**11. Environment, Health & Safety (EH&S) Training:**

Additional training may be required specific to your protocol (hazards you may encounter during the course of performing your job duties.)

<http://www.ehs.ucla.edu> (once there, go to the Training section - bottom right)

**12. For more info, please contact:** Gene Rukavina, DLAM Training Coordinator.

**Email:** [dlamtraining@mednet.ucla.edu](mailto:dlamtraining@mednet.ucla.edu) **Phone/voicemail:** (310) 794-0282

**13. Education & Training FAQs (Frequently Asked Questions):**

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/DLAMEducationFAQs.aspx>

**12. DLAM Class Directions** (must be signed in to the DLAM website):

<https://portal.dlam2.ucla.edu/EducationTraining/DLAMTraining/Pages/DLAMClassDirections.aspx>

**13. View Session Calendar** (must be signed in to the DLAM website):

<https://portal.dlam2.ucla.edu/EducationTraining/Lists/Sessions/AllItemsCal.aspx>

# Reference Email addresses for UCLA- Department of Lab Animal Medicine

Animal Research Committee, ARC Certification, and UCLA policies on animal care: <http://oaro.research.ucla.edu/>

Medical health questionnaire: <https://mhq.healthsciences.ucla.edu/>

Veterinary Staff, Veterinary Technician Staff, Animal Supervisors  
Access Forms, Cage Card Forms, Animal Ordering, Training,  
Pharmacy and Laboratory Services:  
<https://portal.dlam2.ucla.edu/Pages/Default.aspx>

## Important DLAM Numbers:

	<b>Phone</b>	<b>Pager</b>
Dr. Lisa Williams (Clinical Specialist)	794-2455	96667
Dr. Joanne Zahorsky-Reeves (Director of Rodent Medicine)	794-0255	95351
Tensie Palmer (main DLAM number to call)	794-2571	

Training:

Gene Rukavina (Training Coordinator) [dlamtraining@mednet.ucla.edu](mailto:dlamtraining@mednet.ucla.edu)

DLAM Pharmacy

- To place an order for pick up, please use email: [dlampharmacy@mednet.ucla.edu](mailto:dlampharmacy@mednet.ucla.edu)
- Provide the following:  
Principle Investigator  
Recharge ID  
Protocol

Special orders:

**Email is the best way to contact:** [dlampharmacy@mednet.ucla.edu](mailto:dlampharmacy@mednet.ucla.edu)

**Pharmacy Request Form:**

<https://portal.dlam2.ucla.edu/TechOperations/Form Library/Pharmacy Request Form.pdf>

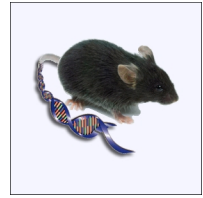
Reference books:

Guide for the Care and Use of Laboratory Animals Eighth Edition (2011): Includes guidelines followed by UCLA as required by AAALAC accreditation. See [http://www.nap.edu/catalog.php?record\\_id=12910](http://www.nap.edu/catalog.php?record_id=12910) for information on ordering. AALAS ALAT, LAT and LATG Training Manuals: [www.aalas.org](http://www.aalas.org)

The AVMA Guidelines for the Euthanasia of Animals: 2013 Edition:

<https://www.avma.org/KB/Policies/Documents/euthanasia-highres.pdf>

# Animal Care Specifics



## **Cage Occupancy Limits**

Lab is responsible for weaning cages and separating over-crowded cages. The lab needs ARC approval to wean at 28 days.

Two females + two litters allowed for only 7 days

**Regular shoe box** Max = 4 mice per cage if greater than 25 grams

**Breeding cages** 2 adults with one litter for 21 days.

**Ventilated cage** Max = 5 mice (greater than 25 grams)

**Food/ Water-** Rodents are given food and water ad lib unless the protocol specifies something different

**Supportive care-** When animals can't reach their food or water, supportive care needs to be given. This includes moistened food pellets on the cage floor, Napa nectar, apple chunks, and even sub-Q fluids if dehydrated.

**Albino/White mice-** Do not put mice with un-pigmented / red eyes on the top shelf in holding rooms. The lack of pigment in their eyes can lead to retinal degeneration, a painful condition that can lead to blindness.

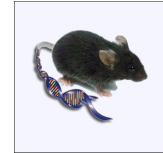
**Mice are escape artists-** Be very careful when handling them. Ensure that the lid is secured properly onto the cage when done handling.

**Allergies-** due to the high content of allergens in the mouse fur, skin and urine, it is very important to follow PPE requirements. Allergies will develop if proper precautions are not taken.

**Dress code-** In addition to proper PPE; long pants and close-toed shoes are required to enter the Vivarium.

**Other information-** Do not open cages outside of laminar flow hood. Fan and light must be turned on and the surfaces sprayed generously with Omega disinfectant before cages can be opened. Do not place food hopper on top of cage top while working with mice

\*These are standard requirements. Always check your protocol for any changes to these rules



# Health Cases

## What happens if an animal gets sick?

- 1) The animal technician will notify the veterinary technician of a sick animal by placing a blue health check post-it on the cage. You may also do this if you find a sick animal and want it to be seen by the DLAM staff.
- 2) The veterinary technician and/or the veterinarian will assess the animal's condition and decide on treatment options.
- 2) You will be contacted about the case. Usually that contact is in the form of an email; for more urgent cases, a veterinarian will call the lab and follow-up with an email. Each email contains information on the location of the animal, a description of its health issue(s), a prescribed course of treatment, information on that treatment (such as how to dilute antibiotics for the drinking water) and a deadline for action.
- 4) Please note that the ARC policy on "Notification of Investigators with Sick or Injured Animals" stipulates that failure to respond to notifications of health reports before the stated deadline and expecting DLAM staff to treat or euthanize the animal is NOT acceptable and constitutes a serious violation reportable to the NIH Office of Laboratory Animal Welfare (OLAW).

If at any time you have any questions, please contact one of the veterinarians whose contact information appears in the email.

## Prescription Medical Treatment

- 1) Please do not begin treatment before getting instructions from the veterinary staff; these will be in the health case email.
- 2) The DLAM Pharmacy can provide you with all medications, etc, that are prescribed. Send requests to: [dlampharmacy@mednet.ucla.edu](mailto:dlampharmacy@mednet.ucla.edu). Place the order before going to the pharmacy so it can be ready for you.
- 3) Treatments and Observations must be accurately documented - it is a medical record. Enter treatments on the orange treatment card on the cage, and initial.
- 4) You should receive an email informing you the treatment has been successful and can be stopped, or the treatment card will be removed by DLAM AHT staff. To stop medical treatment, consult a facility veterinarian.

**Dead animals:** Animals found dead in the cages by technicians will be placed in bags, labeled and stored in the fridge of the facility. Lab will be immediately notified by email. Carcasses may be stored up to 3 days before disposal. Animals should be disposed of properly (bagged and placed in fridge or freezer). Bodies should not be thrown into the trash.

# Injections

## Subcutaneous

Needle size: 25-30g

Amount: 2-3ml maximum

Where: scruff, dorsolateral thorax



## Intramuscular (not shown)

(Not recommended)

Needle size: 27-30g

Amount: 0.05ml/site maximum

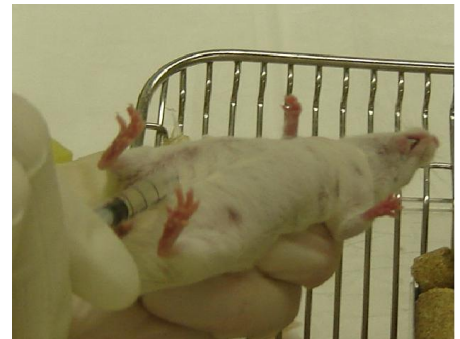
Where: quadriceps, posterior thigh

## Intraperitoneal

Needle size: 25-30g

Amount: 2-3ml maximum

Where: lower right quadrant



## Intravenous

Needle size: 25-30g

Amount: 0.2ml max,  
depending on location

Where: lateral tail vein,  
retroorbital sinus, lateral  
saphenous



# Blood Collection

Total Blood Volume (ml) **1.0 - 2.4**

Safe Bleeding Volume (ml) **0.25**

Total Bleed-out Volume (ml) **0.6 - 1.4**

*The Animal Research Committee (ARC) limits survival blood collection to 1.25% (1.25 ml/100 g) of the animal's current body weight. The frequency of blood collection is dependent upon the volume collected. If the maximum volume is collected, as specified above, blood may be collected once every two weeks. The ARC requires monitoring hematocrit and/or serum protein levels when more frequent collections are necessary.*

Vessel: **Tail vein**

Blood Volume: Up to 10  $\mu$ l

Comment: Accessing the tail vein does not typically require anesthesia. The vein is punctured through the skin with a 25 G needle and blood is collected as it pools on the skin. May be aided by sedation.



Vessel: **Jugular vein**

Blood Volume: Up to 300  $\mu$ l

Comment: Jugular venipuncture is commonly performed under general anesthesia using a 25 G needle attached to a 1-ml syringe.

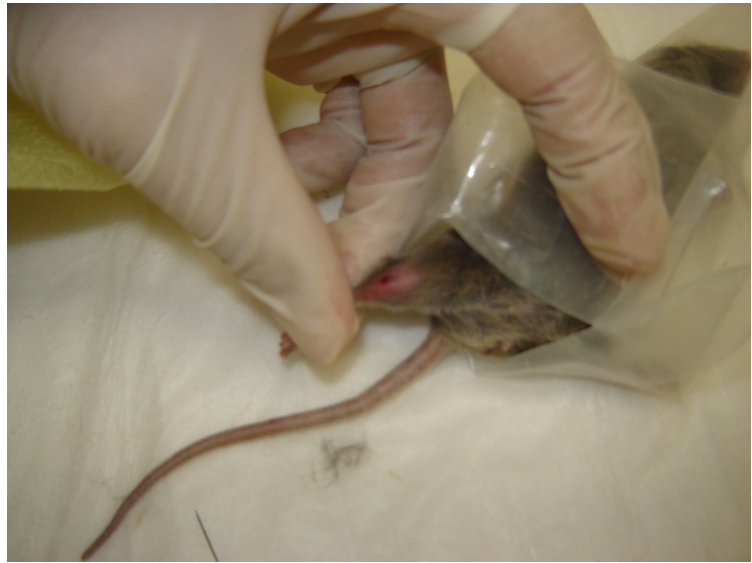
Vessel: **Retroorbital sinus**

Blood Volume: Up to 300  $\mu$ l

Comment: Skilled personnel must perform Retroorbital blood collection under general anesthesia or the risk of injury to the eye and surrounding structures is high. A topical ophthalmic anesthetic (proparacaine) may provide pain relief after the procedure.



Vessel: **Lateral saphenous vein**  
Blood Volume: Up to 50-75  $\mu$ l  
Comment: Accessing the saphenous vein does not typically require anesthesia. The vein is punctured through the skin with a 23 G needle and blood is collected as it pools on the skin. May be aided by sedation.



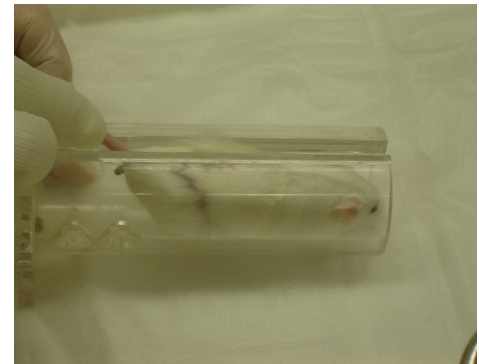
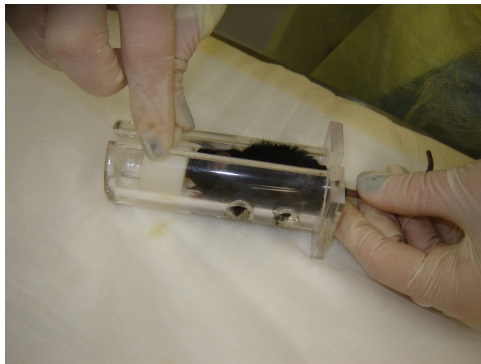
Vessel: **Cardiac puncture**  
(non-survival only)  
Blood Volume: Up to 1.0 ml  
Comment: Cardiac puncture requires general anesthesia. This method is allowed only as a terminal procedure.



# Restraint

A number of commercial restraining devices made of rigid plastic are available for working with rats and can be ordered to correspond with the size/weight.

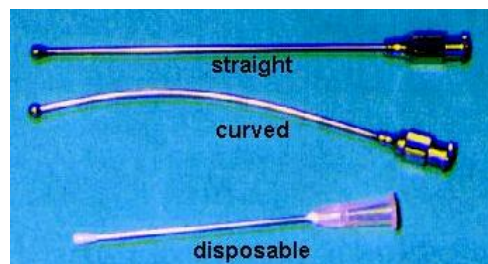
Clear plastic restraint bags are also available commercially (DecapiCones) or can be made out of a freezer-type bag. These bags are in the shape of a cone and have an opening in one corner that the rat's muzzle extends through. These bags are useful for individuals working alone, and injections may be administered through the bag. Animals should be restrained in such bags for short periods of time.



## Oral Gavage

1-2% Body Weight

Oral gavage is the administration of fluids directly into the lower esophagus or stomach using a feeding needle introduced into the mouth and threaded down the esophagus. Feeding needles have a ball tip that makes them atraumatic on delicate oral and esophageal tissues and reduces the chance of introducing the needle into the larynx. Feeding needles are available in a variety of forms and sizes. Reusable needles are entirely made of stainless steel: ball tip, shaft, and hub. Single-use needles have a stainless steel shaft, a silicone ball tip, and a plastic hub. A curved needle is easiest and least traumatic for passage down the esophagus. Lubrication with K-Y jelly should also be used to decrease trauma and to ease passage. Good restraint and head extension are essential to successful placement.



## Biology of the



# Mouse

## Taxonomy

Order: *Rodentia*  
Sub-Order: *Myomorpha*  
Family: *Muridae*  
Sub-Family: *Murinae*  
Genus: *Mus*  
Species: *musculus*



Photo: [www.aalaslearninglibrary.org](http://www.aalaslearninglibrary.org)

## Origin and Habitat

The common house mouse is native to Asia, India, and Western Europe. This species is now found essentially worldwide due to introduction by humans. Mice exist as commensals, living inside during cold weather and moving outside into surrounding fields during the spring and summer, wild forms, living outside throughout their lives, and caged animals, kept for research, testing, teaching, pets and fancy. Although wild mice are nocturnal, commensal and caged mice have periods of activity during both day and night. Commensal mice feed on human food and household items but damage more than they eat. In the wild, mice feed on seeds, fleshy roots, leaves, stems, insects, and some meats, if available. Wild mice are burrowers and build nests wherever suitable cover and food exist.

## Anatomy and Physiology

Adult body weight: 25 - 40 g (female); 20 - 40 g (male) Life span: 1.5 - 3 years Respiratory rate: 94 - 163 breaths/minute Heart rate: 325 - 780 beats/minute Normal average rectal temperature: 99.5°F

- The dental formula is  $2(I\ 1/1, M\ 3/3) = 16$ . The incisors are open-rooted and grow continuously. Mice will bite or "pinch" with their sharp incisors if mishandled.
- The stomach is divided into a proximal nonglandular portion and a distal glandular portion. The two portions are grossly distinct. This is similar to the equine stomach.
- The left lung consists of one lobe, while the right lung consists of four lobes.
- The mouse has five pairs of mammary glands. Distribution of mammary tissue is diffuse, extending from the ventral midline over the flanks, thorax, and portions of the neck.
- Highly concentrated urine is produced; a large amount of protein is excreted in the urine.
- The mouse has the narrowest thermoneutral zone of any mammal thus far measured. A mouse responds to decreases in ambient temperature by nonshivering thermogenesis, and to increases in ambient temperature by decreasing metabolic rate and increasing vascularization of the ears. Nonshivering thermogenesis can produce a 3-fold increase in basal metabolic rate, and for the most part occurs in brown fat. The highest concentration of brown fat is found in the subcutaneous tissues between the scapulae. Brown fat is also called the hibernating gland, although the mouse does not hibernate.
- Bedding should be changed once or twice weekly.

## Nutrition

Mice are fed a commercial pelleted rodent diet and water ad lib. These diets are nutritionally complete and do not require supplementation. Food intake is approximately 1.5g/10g BW/day; water intake is approximately 1.5 ml/10g BW/day.

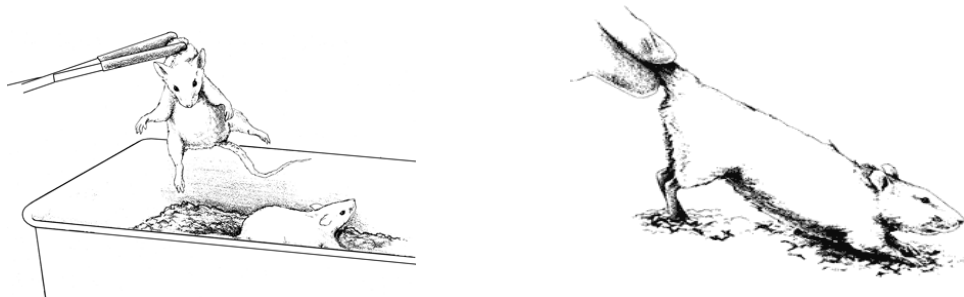
## Uses

Mice are occasionally kept as pets and for fancy. However, the vast majority of caged mice are obtained from laboratory animal breeders for use in biomedical research, testing, and education. In fact, seventy percent of all animals used in biomedical activities are mice. In excess of 1000 stocks and strains of mice

have been developed, as well as hundreds of mutant stocks that are used as models of human diseases. In terms of genetics, the mouse is the most thoroughly characterized mammal.

### Handling

A mouse is lifted by grasping the base of the tail with one's fingers or a rubber-tipped thumb forceps. These are useful techniques for transferring mice from one enclosure to another. To manually restrain the mouse, the mouse is first lifted by the base of the tail, then the loose skin over the neck/shoulder area is grasped between one's thumb and forefinger. This is made easier by lifting the mouse, allowing the mouse to grasp a wire cage top or other surface with the forelimbs, then grasping the skin of the neck/shoulder (scruff) area. With a little practice, the mouse can be lifted and restrained with the one-handed technique. When hand holding, the mouse should be inverted so that the weight of the mouse rests in the palm of the hand. The back end of the mouse is restrained by placing the tail between the handler's fourth and fifth fingers.



### Assisted (with tongs) or Manual (by hand):

Do not ever pick a mouse/rat up by the tip of the tail, only at the base of the tail. It can cause a serious de-gloving (stripping of the skin) injury.

### Handling Pups: Manual - Cup in hands



### Reproduction

Breeding onset is at about 50 days of age in both females and males, although females may have their first estrus at 25-40 days. Mice are polyestrous and breed year round; ovulation is spontaneous. The duration of the estrous cycle is 4-5 days and estrus itself lasts about 12 hours, occurring in the evening. The stage of the estrous cycle can be determined by visual observation of the vulva. Mating is usually nocturnal and may be confirmed by the presence of a copulatory or vagina plug up to 24 hours post-copulation. The presence of sperm on a vaginal smear is also a reliable indicator of mating.

Female mice housed together tend to go into anestrus and do not cycle. If exposed to a male mouse or the pheromones of a male mouse, most of the females will go into estrus in about 72 hours. This synchronization of the estrous cycle is known as the Whitten effect. The exposure of a recently bred mouse to the pheromones of a strange male mouse may prevent implantation (*or pseudopregnancy*), a phenomenon known as the Bruce effect.

The average gestation period is 20 days (19-21 days). A fertile postpartum estrus occurs 14-24 hours following parturition, and simultaneous lactation and gestation may prolong gestation by 3-10 days due to delayed implantation. The average litter size is 10-12 during optimum production, but is highly strain dependent. As a general rule, inbred mice tend to have longer gestation periods and smaller litters than outbred and hybrid mice. The young are called pups and weigh 0.5-1.5 grams at birth, are hairless, and

have closed eyelids and ears. Females should not be disturbed around parturition (from about Day -3 until Day +3) to minimize cannibalism. Pups are weaned at 3 weeks of age; weaning weight is ~10 grams. If the postpartum estrus is not utilized, the female resumes cycling 2-5 days post-weaning.

**Quick Rodent Breeding Information:**

**Weaning Info:** 21 Days; lab needs to separate male and female young into separate cages

**Gestation Info:** duration of pregnancy 19-21 days (Mice); 20-22 days (Rats)

**Sexual Maturity:** 40-60 days (Mice); 65-110 days (Rats)

**Litter Size:** 6-12 Pups (Mice); 7-11 Pups (Rats)

**Best Breeding Time:** 6-8 weeks

**Cannibalism:** First time mothers can cannibalize first litter because pups are ill/ sick or as a survival mechanism for mother or to protect against predators. To help decrease incidence of cannibalism, do not disturb litters for first three days.

**Breeding System Options:** Harem: 1 M + 2-3 F; Monogamous: 1 M + 1 F

**Post Partum Estrus:** female will go into heat (and can be re-impregnated) for 24 hours after giving birth.

**Pseudopregnancy** of ~12-days' duration may follow sterile mating.

**Sex determination pointers in all ages:**

**Male:** Anogenital Distance (distance between anus and genital papilla) is double the length of females, protruding flap of skin of papilla. Males do not have nipples.

**Female:** Anogenital distance is shorter, nipples often seen, hair midline between the anus and genital papilla

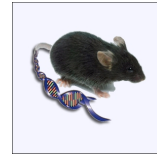
Newborn male mice are distinguished from newborn females by noting the greater anogenital distance and larger genital papilla in the male. This is best accomplished by lifting the tails of litter mates and comparing perineums. Only female mice have nipples.

## Animal Behavior

**Fighting- \*Males are much more aggressive than females, esp. in groups.** Males can only be combined into a group at weaning. Any later, they are likely to fight to the death. Also, if you remove a male from a group cage (such as for breeding), do not put him back in with his brothers, fighting will likely ensue. The aggressor is the one with no wounds. Majority of bite wounds will be at the base of the tail. Separate aggressor to his own cage. Aggression can be inherited, so consider culling aggressors and keeping the other males for inclusion in your breeding program.

**Barbering-** Mice (esp. females) will groom each other to the point where patches of hair loss are evident, esp. on the face/muzzle and back. The mouse without the hair loss is considered the barberer. It is a behavior can be related to stress or disease but is usually considered a normal behavior displaying dominance (aggressor).

# Diseases of the Mouse



## Healthy Animals:

Smooth, well groomed/clean coat, active, curious, eating/drinking, eliminating, interactive, interacting with other mice

## Signs of Illness:

Hunched, ruffled or un-groomed coat, inactivity, lethargic, abnormal breathing patterns, not interacting with other mice; not eating/drinking/defecating/urinating, obvious injury, or tumor growth, study related illness. If you aren't familiar with these signs, watch normal, healthy mice in their cages for comparison.

**Stress**- Happy animals will provide better research results. Stress causes physiological changes that will affect the results of your medical experiments. Manipulation and handling greatly stresses the mice, so handle as efficiently and little as possible. However rats are very social animals and will respond very positively to being gently handled. "Shepherd's shacks" (animal houses), plastic huts or tubes and nestlets are provided for a rodents' emotional enrichment, entertainment and overall well-being. Contact our Pharmacy for assistance in obtaining these items. Stress reduction/ avoidance is mandated by the only federal law that protects animals, the Animal Welfare Act.

## Non-infectious conditions

1. *Barbering* -- Hair nibbling or whisker chewing, is a manifestation of social dominance. Regional alopecia (*hair loss*) around the muzzle may also result from abrasion against cage surfaces
2. *Ulcerative dermatitis*
3. *Tail biting, skin lesions* -- Tail biting and other skin lesions produced by fighting are also manifestations of social dominance. Males tend to be the more aggressive.
4. *Mammary tumors*
5. *Lymphoblastic lymphoma*

## Infectious conditions

- Bacterial, Mycoplasmal and Rickettsial Diseases: *Tyzzler's Disease*: *Bacillus piliformis*, *Transmissible murine colonic hyperplasia*: *Citrobacter rodentium*, *Pseudomoniasis*: *Pseudomonas aeruginosa*, *Pasteurellosis*: *Pasteurella pneumotropica*, *Salmonellosis*: *Salmonella typhimurium* and *Salmonella enteritidis*, *Corynebacteriosis*: *Corynebacterium kutscheri*, *Murine respiratory mycoplasma*: *Mycoplasma pulmonis*, *Erythrozoon coccides*, *Hemobartonella muris*, *Staphylococcus*, *ulcerative dermatitis*
- Viral Diseases: *Mouse Parvovirus*, *Mouse Hepatitis Virus*, *Minute Virus of Mice*, *Sendai Virus*, *Epizootic Diarrhea of Infant Mice*, *Reovirus 3*, *Mousepox*, *Lymphocytic Choriomeningitis Virus*, *Pneumonia Virus of Mice*, *K Virus*, *Lactate Dehydrogenase-Elevating Virus*, *Mouse Cytomegalovirus*, *Mouse Thymic Virus*, *Polyomavirus*, *Mouse Adenovirus*, *Mouse Encephalomyelitis Virus*, *Encephalomyocarditis Virus*, *Murine Leukemia Virus*, *Mammary Tumor Virus*
- Protozoan Diseases: *Toxoplasmosis*: *Toxoplasma gondii*, *Spironucleus (Hexamita) muris*, *Giardia muris*,
- Helminth Infections: *Syphacia obvelata* - pinworm, *Aspicularis tetraaptera* - pinworm, *Hymenolepis nana*, *Hymenolepis diminuta*, *Taenia taeniaeformis*
- Ectoparasites: *Polyplax serrata*, *Myobia musculi* - fur mites, *Radfordia affinis* - fur mites, *Mycoptes musculinis* - fur mites, *Pserergates simplex*,
- Fungal Diseases: *Trichophyton mentagrophytes* - ringworm

# Euthanasia

Euthanasia is defined as a gentle death that is regarded as an act of humane killing with the minimum of pain, fear and distress. It requires proper knowledge of euthanasia concepts, proficient training in methods, and proper animal handling skills

## Approved Procedures – Rodents

- **Never** euthanize animals in the same room where other live animals are present.
- Avoid mixing unfamiliar animals when possible.
- Use **caution** if you must combine cages. **Cage occupancy limits still apply!** (Please refer to page 4 of this document.) Combining male mice from different cages may lead to serious fighting if not euthanized immediately. **Pay close attention to ALL combined cages.**
- Separate all neonates from larger animals.
- Bag all cages for transport to the CO2 chambers, disinfect bags
- Do not leave animals unattended while euthanizing
- CO2 Inhalation
- Check tank pressure. Ensure that connections are secure before beginning.
- CO2 flow must be regulated. **NO DRY ICE!**
- Do **NOT** pre-charge the chamber. Sudden exposure to CO2 at concentrations  $\geq 70\%$  causes distress.
- Place cage in CO2 chamber, open the plastic bag and remove the cage top
- Close and lock the chamber lid before turning on CO2 gas



- Confirmation of death.
  - **Absence of heartbeat must be confirmed!**
  - Not breathing
  - No corneal reflex
  - Blanching/graying of the eyes
  - Sides of the body are sunken in
  - No movement
- If still alive (heartbeat present), repeat CO2 exposure.
- Non- Biohazardous carcasses- Place in a black, trash bag. Secure.
- Biohazardous carcasses- Place into a red, biohazard bag. Secure. Label with medical waste tag.
- Place into a freezer



Pups (1-14 days) must be separated from adults as they can take **up to 20 minutes** to expire.

AVMA Guidelines for the Euthanasia of Animals: After the pups have been deeply anesthetized with CO2 exposure, decapitate them with a pair of very sharp, large scissors for instantaneous death.

Also See ARC Policy: [Euthanasia of Fetuses and Neonates - Mice and Rats](#)

#### Cervical Dislocation

- This method is not permitted within the facilities, without ARC (protocol) approval
- UCLA Policy: Rodents must be anesthetized prior to dislocation
- Requires training and great skill
- Proper use can cause instantaneous loss of consciousness, a painless death and is considered to be humane
- Does not contaminate tissues
- Improper use can cause paralysis and unnecessary distress, suffering and elongated death

## AVMA Guidelines for the Euthanasia of Animals: 2013 Edition



“As veterinarians and human beings it is our responsibility to ensure that if an animal’s life is to be taken, it is done with the highest degree of respect, and with an emphasis on making the death as painless and distress free as possible.”