LABORATORY MICE AND RAT HUSBANDRY

The following link is provided to help you with the Mouse and Rat Lab:

http://www.procedureswithcare.org.uk/category/view-all/

Mice

All laboratory mice used today are bred and raised commercially or in breeding colonies of the user institution. Today, mice are the most widely used vertebrate animal in disease and toxicity testing, as well as in cancer research and biomedical studies. As research animals, mice are economical, easily handled, as well as being the widest range of genetically defined and ecologically refined animals available.

Stocks/Strains-Mice may be classed according to their level of genetic definition as follows:

a) Outbred Stock-Random matings to maintain a constant maximal genetic variation

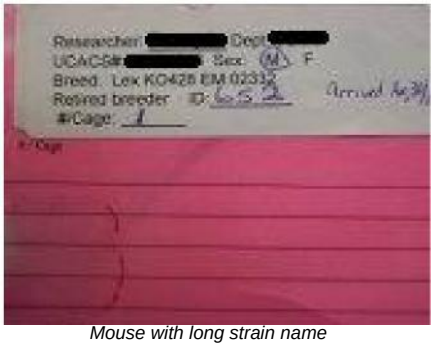
Outbred mouse

b) Inbred Strain-Those which exhibit minimal genetic variation as a result of brother x sister matings for at least 20 successive generations

Balb/c strain of mouse

c) Congenic Strain-Term given to inbred strains into which a single mutant gene has been introduced by a series of back cross matings.

Nomenclature-In the case of genetically defined mice, much of the potential value of the mouse model will be lost if a standardized system of nomenclature and complete record keeping is not observed. Mouse genetic nomenclature is regulated by the Committee on Standardized Genetic Nomenclature for Mice.



Mouse with long strain name

Caging-In one study conducted in mice, vertical dividers were placed in cages, and the animals' performance and well-being compared with that of animals housed in conventional cages. Mice preferred the complex cages, and appeared to be "less emotional" than were the mice kept in regular cages. It was concluded that the divided cage represented a more natural housing arrangement, and that its use would lead to healthier animals. Solid bottom cages with appropriate bedding are strongly recommended for housing rodents, particularly for long-term studies and for breeding rodents. Wire bottom cages, although less labour intensive to use, are far removed from the natural environment. And because of the transfer of heat with stainless steel, the ability of the mouse to maintain its core body temperature may be compromised. Housing breeding animals in wire bottom cages is never recommended. Ideal room temperature for housing mice is 24-25 ° C; mice have the ability to modify their microenvironment by huddling and building nests but maintaining their macroenvironment at this range minimizes stress on the animals.

Mouse in shoebox cage
Large shoebox cage

Ventilated cage rack cage

Microisolator top cage

Cage Density-Mice thrive when housed in groups of two or more per cage. In one study, evidence of "stress" was minimal in mice housed at four per cage, compared with groups of two or eight per cage A high incidence of stress-related tail lesions has been observed in cages housing up to 40 mice which were placed together after weaning. The problem was resolved when the groups were reduced to five per cage. As another example, female C3H/He mice in an intensive breeding program, and housed under conditions of severe social stress, had an incidence of spontaneous mammary tumours considerably different from counterparts kept under ideal conditions.

Several Balb/c mice in a cage

Compatibility is a critical consideration. It may be impossible to house male mice together after puberty, particularly those of more aggressive strains. A common sign of fighting within a cage is the appearance of small "nicks" at the base of tail, often extending down toward the tip, and the tail may be swollen. There may also be scabs along the back of the mouse which aren't clearly visible until the mouse is held for further investigation. Males have been known to fight until death. Females will rarely fight and can be safely placed together at any age without untoward incidents.

Barbering is an expression of dominance behavior seen occasionally in groups of mice of either sex housed together. It is expressed most often as the neat clipping (chewing off) of the facial hairs of cagemates by the dominant mouse. The dominant mouse ends up as the only unshaven member of the community.

Barbered mouse

Artificial Enrichment-Mice have used empty plastic water bottles placed in the cage for a "urinal," and an additional bottle for nesting and as a "bolt hole". It was concluded that provision of the bottle was beneficial in several respects, including improved sanitation, and an opportunity to establish their own optimal environment in the nesting bottles (Boyd, 1988). However, in one study, the addition of objects such as flower pots and bricks has been reported to increase aggression among male mice, presumably because of territorial instincts. Adding paper, wood, pellets or grain to the floor of the cage is another means of environmental enrichment.

Enrichment items

Methods of Identification-Microchips, ear punching, ear tagging, marking tail with permanent marker, dying the fur with non toxic dye, clipping the fur.

Unique Physiological Facts

- Mice do not have sweat glands, heat loss is achieved by decreasing metabolic rate and increasing blood flow to the ears and tail
- Aromatic oils found in softwood shavings (cedar/pine) alter the production of microsomal enzymes in the liver therefore they should never be used as bedding
- Mammary tissue extends onto the back of the mouse therefore mammary tumors may be seen extending over the shoulder area
- Sight is poor, but the mouse has a keen sense of smell and can hear sounds emitted at ultrasonic frequencies

Breeding-The preferred photoperiod for a breeding room is 14 hours daylight and 10 hours darkness. There are three breeding methods used for mice:

1. Monogamous Pair-1 female to 1 male
2. Trio/Triad-2 females to 1 male
3. Harem-2 to 5 females to 1 male

The number of mice that are housed in a cage influence the length of the estrus cycle; singly housed mice will develop abnormally long cycles often with persistent diestrus. The following physiological functions occur in mice that aid in the management of a breeding colony:

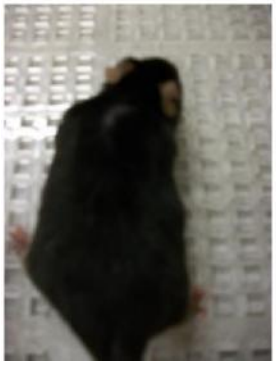
1. The Whitten Effect-anestrous group housed female mice exposed to a male will cycle within 72 hours; females do not need to be in direct contact with a male, sight and smell are enough to induce estrus. This is useful for timed pregnancies.
2. The Bruce Effect-a bred female mouse exposed to strange males/pheromones of a strange male within 48 hours post coitus may block the implantation of the embryos and will return to estrus in 4-5 days. This is important to know as recently bred mice can have blocked pregnancies due to handlers wearing gloves that are contaminated with male urine.
3. The Lee-Boot Effect-females that are group housed without the presence of a male may go into anestrus or have pseudopregnancies (also performing vaginal swabs can induce pseudopregnancy). This state can be relieved by the introduction of male pheromones.

The male will produce secretions from the coagulating glands which aid in forming a vaginal plug after mating. The plug may store sperm, prevent the sperm from leaking out, and may also prevent fertilization by other males in a

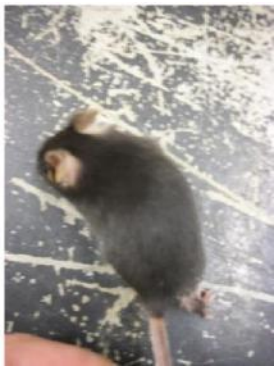
polygamous breeding colony. A vaginal plug found in the cage signals that a mating has occurred within the past 24 hours.

Postpartem estrus occurs 24 hours after giving birth, if the female is mated during this time, there is a 50% success rate that she will become pregnant again.

Signs of Ill Health-Mice are nocturnal, and when several are in a cage they will congregate in a chosen corner. They will rest during the day, moving out occasionally to eat, drink and exercise. A sick animal will exhibit a distinctly different behavior pattern to that of the rest of the group and is usually noticed hunched up, lethargic, and away from the group. Mice have a sleek, shiny coat; if it is roughened and dull, one may suspect the onset of a disease.



Mouse with rough coat



Mouse with sleek coat

Infectious Diseases- Rare due to intensive health monitoring/screening practices of reputable commercial suppliers; **MAY** see Rat Bite Fever, Lymphocytic Choriomeningitis (LCM) and ringworm. If housing wild-caught mice safety measures must be undertaken for Hantavirus.

Brief Overview of Transgenic Mice

Over the past twenty years or so, the confluence of embryology and genetic engineering has resulted in the creation of transgenic animals; animals whose genome has been deliberately modified. For practical reasons, i.e., their small size and low cost of housing in comparison to that for larger vertebrates, their short generation time, and their fairly well defined genetics, mice have become the main species used in the field of transgenics.

A representative, but non-inclusive, list of purposes for which transgenic animals have been used indicates the wide ranging application of this biotechnology:

- in medical research, transgenic animals are used to identify the functions of specific factors in complex homeostatic systems through over- or under-expression of a modified gene (the inserted transgene)
- in toxicology: as responsive test animals (detection of toxicants)
- in mammalian developmental genetics
- in molecular biology, the analysis of the regulation of gene expression makes use of the evaluation of a specific genetic change at the level of the whole animal
- in the pharmaceutical industry, targeted production of pharmaceutical proteins, drug production and product efficacy testing
- in biotechnology: as producers of specific proteins
- genetically engineered hormones to increase milk yield, meat production; genetic engineering of livestock and in aquaculture affecting modification of animal physiology and/or anatomy; cloning procedures to reproduce specific blood lines
- developing animals specially created for use in xenografting.

Laboratory Animal Management

Physical and biological containment for transgenic animals should be adequate to assure the biosafety of the animal care staff which work with the animals, to prevent any possibility of the transfer of the gene within the non-transgenic colonies maintained in the same facilities, and to protect potentially immuno-compromised transgenic animals from pathogens.

Establishing and maintaining lines requires careful management. As part of the general process of transgenic animal creation, each animal used must be carefully identified. Cage cards and good records with details of

breeding information are necessary to be able to identify with certainty the genetic characteristics and modifications of the animal. The data recorded should include the identity, breeding, pedigree and any other pertinent data such as any dates, observations or laboratory analysis information. Since transgenic animals are not easily replaceable, the cost of containment is an important factor in transgenic experimental design.

Embryo freezing is used for the preservation of transgenic strains. To protect colonies against disease, contamination or any other cause of loss, a large number of preimplanted embryos are kept, by cryopreservation. This also reduces the cost of maintaining a transgenic mouse line when it is not needed for experimentation.

With the development of transgenic animals, just a small genomic change can induce unpredictable and quite drastic changes at the level of the whole animal. This is the main challenge for transgenic animal management. It is therefore important to have a clear procedure for monitoring the animals and for dealing with unanticipated suffering.

Transgenic mice

Rats

The development of today's laboratory rat has essentially been an American initiative and the great majority of the strains used today have originated in the U.S.A. The laboratory rat accounts for at least 20% of the total number of laboratory mammals used. They are used in every aspect of biomedical and behavioral research and testing.

Interesting Note: In employment situations involving sensitization to laboratory animal dander, the most frequent allergen source appears to be the rat.

Stocks/Strains- Like mice, rats are available in various genetically defined varieties but are much less numerous and often much less well defined than the mouse strains.
The three most common outbred stocks of rats are:

1. Sprague-Dawley: average size white rat
2. Wistar: average size white rat but with a wider head and shorter tail then the Sprague Dawley
3. Long-Evans: a "hooded" rat that is smaller than the other two

Caging- Rectangular, solid plastic cages at least 20 cm high with a wire mesh lid as rats like to stand up and scan their surroundings.

Rats in cages

Cage Density-Rats are frequently housed singly for certain types of studies; however, it is desirable that two or more compatible rats be housed together in an appropriate cage. Post-puberal males are usually compatible, particularly if they have been together since an early age. It has been shown that even groups of highly standardized male rats exhibit a high level of variability of behavioural patterns.


Several rats in a cage

Artificial Enrichment-Rats be supplied with appliances such as PVC plastic pipe to use as a means of burrowing and to use as a "bolt hole". Adding paper, wood, pellets or grain to the floor of the cage is another means of environmental enrichment.


Enrichment items

Methods of Identification- Microchips, ear punching, marking tail with permanent marker, dying the fur with non toxic dye, clipping the fur.

Unique Physiological Facts

- Rats also do not have sweat glands, heat loss is achieved by decreasing metabolic rate and increasing blood flow to the ears and tail
- They do not have gall bladders; the pancreas aids with digestion by the numerous ducts that lead directly to the small intestine
- Rats do not have the ability to vomit
- Rats are monogastric omnivores which is why they are able to survive in different types of habitats.
- If housed by themselves, rats will incur marked changes in their disposition, adrenals, thyroids, and microsomal liver enzymes; and such behavioral parameters such as alcohol consumption
- The overproduction of porphyrin pigments by the Harderian gland (located behind the eyes) is a non-specific response to stress, pain, disease, or poor diet. The porphyrin manifests itself as red staining around the animal's eyes and nose, and is often mistaken for blood. This shedding of "red tears" is known as chromodacryorrhea.


Porphyrin staining around nostrils

- Aromatic oils found in softwood shavings (cedar/pine) alter the production of microsomal enzymes in the liver therefore they should never be used as bedding

Breeding-Rats breed year round; litter frequency will decrease in winter months unless artificial light is used to maintain a 14 hour photoperiod of light. The same three breeding methods used in mice can also be used with rats:

1. Monogamous Pair-1 female to 1 male
2. Trio/Triad-2 females to 1 male
3. Harem-2 to 5 females to 1 male
4. Other breeding system such as rotating 1 male between 7 separately caged females, allowing one week with each immediately following weaning

Timed pregnancies are accomplished by overnight pairing or a two hour pairing on the morning that a vaginal smear shows positive for estrus. Estrus may be synchronized using the Whitten Effect as outlined in the mouse section.
If utilizing post partum breeding, the time from fertilization to birth may be lengthened to 30 or more days due to delayed implantation which is proportional to the number of young being suckled by the female. Failure to conceive at post partum estrus will delay breeding until two to four days after the litter is weaned.
Sexing of pups can be done on neonates by comparing the ano-genital distances between littermates, with the distance in the male being twice that of the female. In addition, the male genital papillae are larger.

Signs of Ill Health- As with mice, a sick rat will exhibit a distinctly different behavior pattern to that of the rest of the group and is usually noticed hunched up, lethargic, and away from the group (if group housed), or isn't inquisitive when a person enters the room. They also have a sleek, shiny coat and if it is roughened and dull, one may suspect the onset of a disease.

Infectious Diseases- Rare due to intensive health monitoring/screening practices of reputable commercial suppliers; **MAY** see Rat Bite Fever and ringworm.

Physiological and Nutritional Parameters on Mice and Rats*

SPECIES	Rectal Temp. °c +0.5	Resp. Rate/ Mean and	Heart Rate/ Mean and (range)	Average Daily Water Consumption	Urine Excreted Daily	Daily Feed Recommendations	Digestible Protein**
MOUSE	37.5	138 (94-163)	470 (325-780)	3-7 ml	1-3 ml	3-6 g	12
RAT		92 (70-115)	350 (250-450)	20-45 ml	10-15 ml	10-20 g	12

* Averages and ranges derived from literature mean values for **young adult animals** under various conditions (from various sources).
** Refers to (ideal or digestible protein required; crude protein (CP)) levels in most prepared laboratory animal diets may be considerably higher.

Breeding and Reproduction Data on Mice and Rats:

SPECIES	Breeding Age range	Cycle Type Length	Duration of Sexual Receptivity	Breeding Behaviour and Season	Gestation Range	Litter Size and Rang	Optimal Reproductive Span
MOUSE	6 weeks	4-5 days Polyestrous	10-20 hours	Harem (1♂ to 4♀) All year	19-21(20	6-12	7-8 months
RAT	10-12 weeks	4-5 days Polyestrous	10-20 hours	Harem (1♂ to 6♀) All year	20-22(21	7-14	9-10 months