IACUC Approved on 8/16/2023

# **University of Southern California**

# Institutional Animal Care and Use Committee Rodent Housing, Breeding and Overcrowded Cage Policy

## A. Background

To provide information to USC researchers and animal care staff regarding proper housing and breeding practices for mice and rats, and how to identify and address overcrowded cages.

#### **B.** Definitions

Harem breeding: One male with three or four females

<u>Pair breeding</u>: One male with one female <u>Trio breeding</u>: One male with two females

#### C. Applicability

The following housing, breeding and overcrowding policies are applicable to all mice (*Mus musculus*) and rats (*Rattus norvegicus*) housed in USC animal facilities.

The laboratory personnel who manage the colonies are expected to maintain breeding records and keep up with colony management. While animal care staff play a key role in checking the animals and alerting researchers about overcrowded cages, it is the researcher's responsibility to house the animals appropriately and wean on time.

Overcrowded cage reminders are a courtesy to ensure compliance, but they should not be routine or solely relied upon for colony management.

#### D. Policy

#### General Housing:

The maximum number of adult mice per cage is 5, and the maximum number of adult rats (up to 500 grams in weight) per cage is 2.

- Very large rats weighing >500 grams must be single housed.
- This is based on the average weight of adult mice and rats, the size of caging used at USC, and recommended cage spaces outlined in the *Guide for the Care and Use of Laboratory Animals* (see Appendix 1).

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Male mice should be housed together from the time of weaning due to their aggressive and territorial nature.

- It is a welfare concern to mix male mice from different litters in the same cage due to the high risk of fighting and injury.
- Mixing males from different litters to reduce per diems is not allowed or justifiable, and can result in non-compliance.
- Once a male mouse has been used for breeding, he should remain either single housed or with his female mate(s) and not put back together with other males.

The standard weaning age for mice and rats is **21 days old**, unless otherwise approved in the IACUC protocol.

- Please contact the veterinary staff (<u>vetmed@med.usc.edu</u>) if there is a concern that a particular strain of pups are consistently too small to wean at 21 days.
- When weaning mice, the researcher(s) must add food pellets and ensure the
  mice have a water source (i.e. water valve or water bottle depending on the
  facility).

## Breeding:

Pair breeding is the standard practice at USC.

- Only one male should be housed in a breeding cage.
- For mice, trio or harem breeding may be permitted with acceptable justification and prior IACUC approval.
  - Additional rules for trio or harem breeding apply (see Appendix 2 and 3).
- Researchers are responsible for maintaining their own breeding records.
   Maintaining good records will help to avoid consistent overcrowded notices and possible service fee charges.

Actions required when a cage is found improperly housed (>1 male with a female past weaning) or an improper breeding scheme is being followed:

- Animal care staff will notify the contact on the cage card and copy the PI and veterinary staff.
- Animal care staff may separate the cage immediately without notice and a service fee will be applied to avoid animal welfare concerns that may result from housing multiple males with females (aggression, wounds, unwanted breeding).
   This charge will incur for all future incidences of improper housing as well.

#### Overcrowding:

A cage is considered overcrowded or improperly housed in the following situations:

- More than 5 adult mice are present in the cage past weaning.
- More than 2 adult rats >200 grams are present in the cage.

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- o A female rat with a litter is considered a full cage.
- The male rat must be removed when the female has a litter. We recommend doing so when the female is around two weeks pregnant to avoid stress.
- More than 1 adult rat >500 grams is present in the cage.
- More than one adult male is in a cage with a female past weaning.
  - Note: This is never an acceptable housing scheme, as it leads to fighting and aggression between the males and is an animal welfare concern.
- There is more than one litter of pups, and the total combined number of pups is 12 or more.
- There are multiple litters born more than 6 days apart.

## Actions required when a cage is found overcrowded:

- Animal care staff will notify the contact person listed on the cage card via email.
- If there is not an immediate animal welfare concern, then the contact person has 48 hours from the date of the email notification to separate and properly house the animals.
- If the cage is not separated and properly housed in 48 hours, then the animal care staff will separate the animals and a service fee will be applied.
  - It is not acceptable to mark the cage for euthanasia and leave it overcrowded.
  - It is not acceptable to pool animals from multiple cages into one cage to be euthanized.
  - It is not acceptable to remove the overcrowded card unless the cage is no longer overcrowded according to the standards outlined in this policy.
- If the overcrowded cage is an immediate animal welfare concern, then the animals will be separated immediately and the PI will be charged a service fee will be applied.
  - Examples of this include trampling, fighting, increased humidity/moisture in the cage due to severe overcrowding, or a sick case reported as a result of the housing scheme.

#### E. References

 Committee for the Update of the Guide for the Care and Use of Laboratory Animals, et al. (2011). Guide for the Care and Use of Laboratory Animals. 8th ed., National Academies Press. https://www.ncbi.nlm.nih.gov/books/NBK54050/

## F. Appendix

1. Minimum floor space requirements for group-housed laboratory rodents (via the *Guide*, 2011).

TABLE 3.2 Recommended Minimum Space for Commonly Used Laboratory Rodents Housed in Groups\*

Animals	Weight,	Floor Area/Animal, <sup>a</sup> in. <sup>2</sup> (cm <sup>2</sup> )	Height, <sup>b</sup> in. (cm)	Comments
Mice in groups <sup>c</sup>	<10 Up to 15 Up to 25 >25	6 (38.7) 8 (51.6) 12 (77.4) ≥15 (≥96.7)	5 (12.7) 5 (12.7) 5 (12.7) 5 (12.7)	Larger animals may require more space to meet the performance standards.
Female + litter		51 (330) (recommended space for the housing group)	5 (12.7)	Other breeding configurations may require more space and will depend on considerations such as number of adults and litters, and size and age of litters.
Rats in groups <sup>c</sup>	<100 Up to 200 Up to 300 Up to 400 Up to 500 >500	17 (109.6) 23 (148.35) 29 (187.05) 40 (258.0) 60 (387.0) ≥70 (≥451.5)	7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8) 7 (17.8)	Larger animals may require more space to meet the performance standards.
Female + litter		124 (800) (recommended space for the housing group)	7 (17.8)	Other breeding configurations may require more space and will depend on considerations such as number of adults and litters, and size and age of litters.
Hamsters <sup>c</sup>	<60 Up to 80 Up to 100 >100	10 (64.5) 13 (83.8) 16 (103.2) ≥19 (≥122.5)	6 (15.2) 6 (15.2) 6 (15.2) 6 (15.2)	Larger animals may require more space to meet the performance standards.
Guinea pigs <sup>c</sup>	Up to 350 >350	60 (387.0) ≥101 (≥651.5)	7 (17.8) 7 (17.8)	Larger animals may require more space to meet the performance standards.

<sup>\*</sup>The interpretation of this table should take into consideration the performance indices described in the text beginning on page 55.

#### 2. Rules for trio breeding in mice:

a. No more than 12 pups are allowed in a cage (unless all from the same litter).

<sup>&</sup>lt;sup>a</sup>Singly housed animals and small groups may require more than the applicable multiple of the indicated floor space per animal.

<sup>&</sup>lt;sup>b</sup>From cage floor to cage top.

<sup>&</sup>quot;Consideration should be given to the growth characteristics of the stock or strain as well as the sex of the animal. Weight gain may be sufficiently rapid that it may be preferable to provide greater space in anticipation of the animal's future size. In addition, juvenile rodents are highly active and show increased play behavior.

<sup>&</sup>lt;sup>4</sup>Other considerations may include culling of litters or separation of litters from the breeding group, as well as other methods of more intensive management of available space to allow for the safety and well-being of the breeding group. Sufficient space should be allocated for mothers with litters to allow the pups to develop to weaning without detrimental effects for the mother or the litter.

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- b. If there are 2 litters in a cage, they must be less than 6 days apart and have no more than 12 pups combined.
- c. If one female has pups, the other female must be removed (if pregnant) before she has pups.
- d. If both females have pups, the female with the older litter must be moved to a separate cage.

## 3. Rules for harem breeding in mice:

- a. Pregnant females will be removed from the cage before giving birth.
- b. No pups will be born in a harem breeding cage.
- c. If a female has pups within the harem cage, the other adult mice must be moved to another cage.