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1. Purpose

Drexel University Institutional Animal Care and Use Committee (IACUC) established this document to provide information about policies and best practice recommendations for the proper maintenance of rodent breeding colonies, specifically mice (*Mus musculus*) and rats (*Rattus norvegicus*).

2. Definitions

Pair breeding: Breeding of one (1) adult male and one (1) adult female.

Trio breeding: Breeding of one (1) adult male and two (2) adult females.

Harem breeding: Breeding one (1) adult male and three (3) adult females.

Post-partum estrus (continuous breeding): Female rodents may undergo a fertile estrus 14-24 hours following parturition. Females that are housed with males when they give birth can therefore become pregnant with a second litter that would be delivered around the time of the weaning date of the first litter. This can lead to overcrowding if the weanlings are not separated at 21 days of age.

Female/Female co-parenting: Two (2) females in a cage rearing a litter.

Litter: Group of pups

Pup: Neonatal rodents up to 21 days of age

Weanling: Young rodent recently separated from its mother; usually 21 - 23 days old.

Weaning: The moment pups are transferred out of the mothers' cage.

Delayed weaning: Weaning >21 days.

Overcrowding: For mice, overcrowding occurs when there are more than 5 adult mice per mouse cage after weaning. In other species, such as rats, overcrowding is determined according to the weight of the animal in relation to the size of the cage.

Delayed weaning: Weaning >21 days.**Breeding or Newly Weaned Cage Tags:** Additional accessory cage tag provided in the back of the cage card as cage-side communication regarding the breeding or weaning status of the cage.

⚠: **REQUIREMENT** per this procedural document

★: **BEST PRACTICE RECOMMENDATION (**optional but suggested)

3. IACUC Protocol Requirements

⚠ All breeding procedures must be described and justified in an approved IACUC protocol. Any deviations or changes from the breeding scheme or age of weaning must be approved in an amendment submitted to the IACUC prior to implementation of any of these changes. Amendment forms are submitted through DragonSpot.

3.1 Protocol Information

a. Section E: Include predicted numbers of animals generated based on breeding strategy, predicted numbers of animals that can be used for experiments based on genotypes needed (keeping in mind whether predicted genotypes will be generated according to Mendelian genetics). The IACUC can provide a template Excel file for complicated breeding sequences that may provide guidance with calculating the predicted number of animals generated.

b. Section H: Breeding scheme, weaning age, approximate duration that breeders are used, method for obtaining tissue samples for genotyping, method for identifying pups (ear punch, ear tags, or tattoo)

4. Breeding Schemes

4.1 Pair Breeding:

a. This is the standard practice for breeding.

b. One male (only one male per cage is allowed) and one female are housed together for mating.

c. Mice: Continuous breeding should be used unless otherwise approved and justified in an IACUC protocol. Male and female breeders do not need to be separated when the female becomes pregnant or delivers the pups

1. This model takes advantage of postpartum estrus and allows female to become pregnant and nurse at the same time. Litters are born approximately 21 days apart.
2. ⚠ **Provisions to ensure cages do not become overcrowded must be in place,** which includes weaning on time (see Weaning section). Simultaneous presence of two or more litters in a single cage is not permitted, unless scientifically justified and approved in the IACUC protocol (see Housing Compliance and Overcrowding in Section 7).

   a. Keeping an older litter with a newborn litter endangers the health of the newborn pups:

      1. The older pups will overcrowd the cage and can trample the newborn pups
      2. The older pups may compete with the newborn pups for resources (e.g., nutrition from the dam)

   3. ⚠ **Note:** If breeding male mice are separated, they must be singly housed due to their aggressive nature after breeding. Breeding males may cause grave physical harm or death to a male cage mate if cohoused again. Alternatively, the male may be placed in another breeding cage.

d. ⚠ **Rats:** The male must be removed when the female is noted to be pregnant (recommended) or sometime prior to giving birth, as a female and a litter is considered a full cage.

4.2 Trio or harem breeding:

a. ⚠ Must be approved in an IACUC protocol. Justification must be provided for this increased density, including a description of management techniques to minimize detrimental effects on the mother and litters to maximize pups' ability to develop to weaning age.

b. ⚠ Females must be separated when noted to be pregnant unless otherwise approved and justified in an IACUC protocol

1. ⚠ **Rats:** females must be singly housed when noted to be pregnant.

2. ⚠ **Mice:** female mice must either be singly housed when noted to be pregnant or one female can be left with the male for continuous pair breeding.

a. If the protocol is approved to leave two (2) pregnant female mice together to co-mother/co-nurse, **the females must be separated if there are more than 12 pups present between both litters or if the pups are > 5 days apart (see Section 7, Housing Compliance and Overcrowding).** It's recommended to separate the second female if she has not given birth or is not obviously pregnant by day 5 after first litter is born, to prevent litters >5 days apart. The male must also be separated in this scenario unless continuous trio breeding is approved and justified in IACUC protocol.

c. After pups are weaned, the females may be returned to the original breeding scheme.

4.3 Retirement of Breeders

a. ★ **Best practice recommendation:** Fertility of rodents is highest during the first 6 months of age. After a year of age, fertility is very low. Therefore, the IACUC recommends retiring breeders between 8 to 12 months of age. Individual genetic lines will vary, and investigators must use their best judgment and experience to decide when to replace breeders.

5. Pregnancy and Birth

5.1 Monitoring for Pregnancy and Birth

a. The designated colony manager has the primary responsibility for checking for pregnancy and keeping accurate and detailed breeding records.

b. The lab is responsible for monitoring and separating of pregnant females (if needed), according to protocol approved procedures.

c. ★ **Best practice recommendations:** It's recommended to not change the cage if the female appears heavily pregnant, unless required. If required to do so, it's recommended to transfer some dirty bedding and the nest for familiar scents and to decrease stress around parturition.

5.2 After Birth

a. ⚠ When the litter is born, the date of birth (DOB) and projected weaning date must be documented on the breeding cage tag (see Appendix 1).

1. This is the responsibility of lab personnel. However, University Laboratory Animal Resources (ULAR) personnel will note the DOB on the breeding cage tags when litters are found during routine cage checks and changes.

b. ★ **Best practice recommendations:**

1. After pups are born, leave the cage undisturbed for at least 5-7 days, except for replenishing food and water as needed.

2. If the cage must be changed, recommend following these practices:

a. Transfer the female to the clean cage first.

b. Transfer the litter, a small amount of dirty bedding, and the nest all together to the new cage with the female.

6. Weaning

6.1 Weaning age

a. ⚠ All litters must be weaned by 21 days of age, unless otherwise approved and justified in protocol.

1. If day 21 falls on a Saturday and lab staff is not available to wean on the weekend, pups should be checked for weaning readiness on Friday (i.e., day 20) and wean if the pups are of appropriate size (~10 grams). If day 21 falls on a Sunday, and lab staff is not available to wean, pups should be weaned on Monday (i.e., day 22). If pups are not of adequate size, they should be checked daily for weaning readiness.
2. Provisions must be in place to maintain breeding colonies over extended holidays or breaks, to prevent overcrowding.

b. ⚠ In the case of some inbred, genetically modified, or mutant strains, it may be advantageous to allow pups to remain with the female for 28 days. This delayed weaning must be approved in the IACUC protocol. It is recommended to submit an amendment to IACUC protocol if this is routinely observed in a strain (with veterinary approval, see below).

1. The breeding cage tag should be updated with the 28-day weaning date in cases where this is approved by the IACUC or veterinary staff.
2. Even with prior IACUC approval for 28-day weaning, cages should still be checked for weaning readiness at 21 days of age to prevent overcrowding, especially in cases of continuous breeding or trio breeding where the females are approved to stay together.

c. As a general rule, mouse? pups should be at least **10 grams** at weaning.

1. ⚠ Any mouse pups unexpectedly weighing less than 10 grams at 21 days must be reported to veterinary staff for a health evaluation and to extend weaning age, if the protocol does not have approval for up to 28-day weaning. If any other animal care concerns are noted, please contact ULAR for additional guidance and assistance. Supportive care (diet gel and hydrogel) is recommended before and after weaning to help pups grow.
2. **⚠ If a new litter is born and the older litter is not yet ready to be weaned (due to small size), ULAR veterinary staff must be contacted for further guidance.**

d. A "Weaning Calendar" is provided in Appendix 2 of this document and within the Office of Research and Innovation's Guidelines and Procedures Catalog.

6.2 Weaning Practices

a. ⚠ Pups must be separated by sex into individual male or female cages, keeping in mind that housing density should not exceed 5 mice/cage and 2 rats/cage (depending on weight – see Housing Compliance and Overcrowding in Section 7).

1. It is not acceptable to mix male mice from different litters, as this can lead to increased risk of aggression.

b. Place a "Newly weaned" cage tag and include a date that is 2 weeks from date of weaning.

c. ★ **Best practice recommendations:** Provide supportive care for the first 2 weeks after weaning, such as hydrogel and/or a water bottle with long sipper tube. Food pellets should also be provided on the floor of the cage at weaning.

d. If a lixit is present, the lixit should be toggled at the time of weaning.

7. Housing Compliance and Overcrowding

7.1 Housing Guidelines

a. Refer to the *Guide for the Care and Use of Laboratory Animals* (also in Appendix 3) for specific guidance on cage size and allowance of number of animals. Generally, a cage is considered overcrowded or improperly housed when:

- More than 5 adult mice are present in the cage past approved weaning period
- Rat cage size allowances
  - Up to 300g per rat = 29 sq inches per rat= 4 rats
  - Up to 400g per rat = 40 sq inches per rat = 3 rats
  - >to 500g per rat = 60 sq inches per rat = 2 rats
  - A female rat with a litter is considered a full cage
  - The male rat must be removed when a female has a litter.
- More than 1 male is present in a cage with a female past weaning
- More than 1 litter of pups where the total combined number of pups is 12 or more
- Multiple litters more than 5 days apart

7.2 ⚠ Overcrowding Non-Compliance

a. Note, ULAR staff is not responsible for notifying of weaning dates. It's the lab's responsibility to monitor, document, and wean on time. Laboratory personnel are responsible for monitoring their breeding colony, including on weekends and holidays.

b. If overcrowding is noted by ULAR staff or if a litter is overdue for weaning, an **"Overcrowded" card** will be posted on cage by ULAR with the current date noted on the card.

1. For routine/overdue weaning, overcrowding or non-compliance with breeding scheme where animal welfare is not at immediate risk: The lab is responsible for weaning and/or separating the animals within **2 calendar days** (inclusive of weekends and holidays) of the posting. ULAR will no longer be sending e-mail notifications of overcrowding, unless there is an animal welfare concern. If the cage(s) is not addressed within 2 calendar days, ULAR husbandry staff will wean and/or separate the animals on the third day after the overcrowded card is placed. The PI will incur a **$50 charge per newly weaned cage** for the technical staff providing If a breeding strategy results in two litters from the same dam the older litter must be weaned to prevent overcrowding and trauma to the newborn pups. Cages identified with two or more litters will be reported to the PI. If the younger litter appears to be unaffected by the presence of the older pups, the investigator must wean or separate the older litter within **2 calendar days (including weekends and holidays), but ideally sooner if possible.** If the younger litter is being harmed by the older pups, ULAR will immediately wean the older litter, a $50 charge per newly weaned cage will apply, and the investigator will be notified.

Note: if the older litter is not yet ready to be weaned, please contact veterinary staff for recommendation.

2. Repeated instances of overcrowded status (defined as 3 incidences of ULAR-charged weaning) will result in the PI being considered non-compliant with this policy. Before overcrowding escalates to a non-compliance, ULAR may request the PI and/or applicable laboratory personnel to complete targeted retraining on rodent breeding and colony management procedures to prevent recurrence and non-compliance. Non-compliance can be prevented with regular consultation with ULAR regarding any breeding issues noted in your colony, and timely weaning of animals as described in the approved IACUC protocol and this policy. Every effort should be made to resolve breeding related issues prior to reporting these issues to the IACUC.

8. Cage Identification

a. All breeding cages MUST be marked with a breeding tag provided by ULAR.
   1. The breeding tag should include breeding set up date, date of birth of the litter, and wean date.
   2. When animals are newly weaned, the "Newly weaned" tag is placed on the cage to identify them in the first 2-weeks post-weaning in case additional support or monitoring is warranted.

b. Investigators may create their own breeding cage cards, but breeding tags should still be used as cage-side communication with ULAR staff to prevent their disrupting new litters when changing cages.
   1. If the PI/staff chooses to make breeding cards, the following information must be included:
      a. Protocol Number
      b. Name of PI
      c. The date the breeding cage is set up
      d. Date of birth of the litters
      e. Anticipated weaning date.

9. Breeding Reports

All in-house births, including all embryos greater than E16 and all births (live and stillborn), even if the animals are euthanized prior to weaning, must be reported monthly to ULAR for maintaining approved animal numbers on each IACUC protocol. An In-House Birth Report must be submitted monthly, even if no animals were produced during the month. The report should be submitted by the 5th of the following month. The In-House Birth Report is an online form and can be accessed using the link provided: In-House Birth Report Form and is available on the Animal Care and Use webpage under "Animal Care and Use Procedures". Any questions can be sent to ULAR@drexel.edu.

10. Responsibilities

10.1 Drexel University IACUC Responsibilities

The Drexel University IACUC and the IACUC Office are responsible for maintaining this guidance document, training, and monitoring. All exceptions to this policy must be approved by the IACUC. For inquiries regarding these procedures, please contact the Director of Animal Welfare, on behalf of the Office for Research & Innovation (ORI), or the Attending Veterinarian, as part of University Laboratory Animal Resources.

10.2 Principal Investigator Responsibilities

Principal investigators (PI) and their designated staff who are managing rodent breeding colonies under an approved IACUC protocol are required to adhere to this policy.

It is the PI's responsibility to plan colony management, including, but not limited to timely weaning of litters and prevention of overcrowded cages. PIs must ensure that an efficient communication method is in place for notification from husbandry and veterinary staff regarding animal issues.

11. Resources

Strategies for Maintaining Colonies of Laboratory Mice: A Jackson Laboratory Resource Manual Ray Lambert (Sr. Editor) 2009.

The Guide for the Care and Use of Laboratory Animals, 8th edition.

12. Revisions

Edition 001/Effective Date: 3/2/2016 - Original Document

Edition 001/Review Date: 09/2021

Edition 002/Revision and Effective Date: 05/22/2023 - Revised Document.

Updated Mouse Breeding Policy to become a Rodent Breeding Policy to include rats, *Rattus rattus.* Revision to breeding report submission procedures. Added Weaning Calendar.

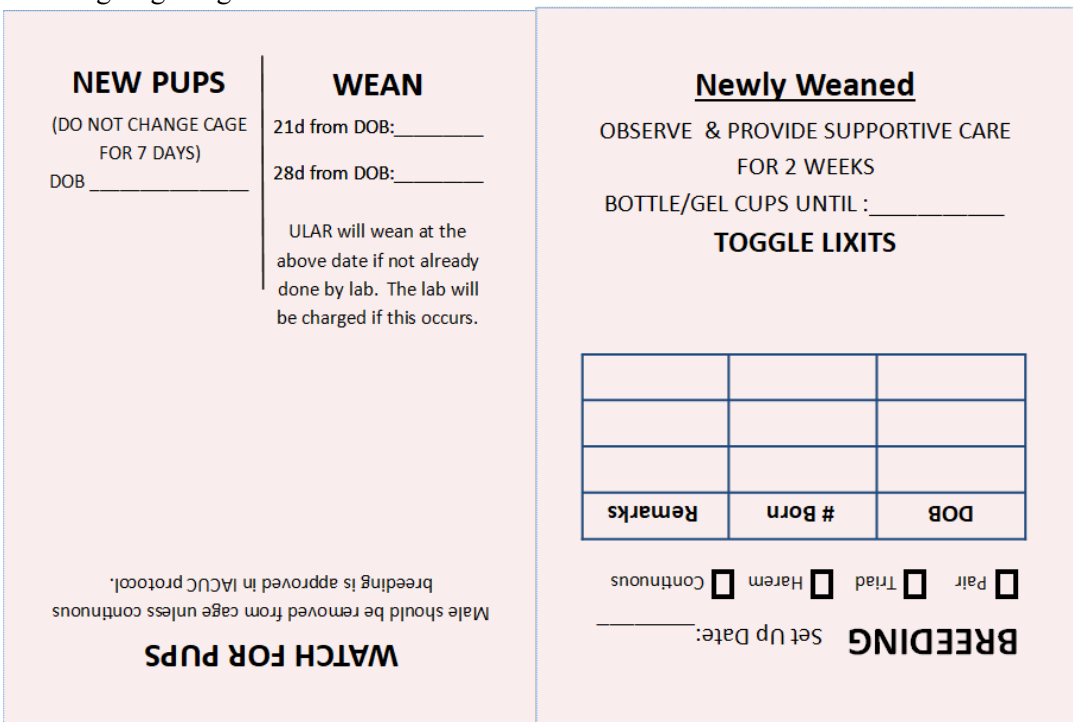
Edition 003/Revision and Effective Date 6/10/26

1. For mice, continuous, pair breeding is now the standard breeding scheme.
2. Addition of separation guidance for trio and harem breeding to avoid overcrowding
3. Removal of the 21-23 days of age to wean and replaced with 21 days of age to avoid overcrowding
4. Rat cage size/population guidance
5. Addition of a charged fee ($50/cage) when ULAR weans animals (two days past weaning date)
6. Guidance for when two litters are present in the same cage: Older litters must be promptly weaned when co-housed with newborns; if no harm is seen, separate within

2 days, but if harm occurs, ULAR will immediately wean (with fee) and notify the PI. Consult veterinary staff if weaning timing is a concern.

Appendix 1

Breeding Cage Tags



Appendix 2

21 Day weaning Schedule

JANUARY		FEBRUARY		Leap Year	MARCH		APRIL		MAY		JUNE		JULY		AUGUST		SEPTEMBER		OCTOBER		NOVEMBER		DECEMBER	
DOB	Wean on (21d)	DOB	Wean on (21d)	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)	DOB	Wean on (21d)
1/1	1/22	2/1	2/22	2/22	3/1	3/22	4/1	4/22	5/1	5/22	6/1	6/22	7/1	7/22	8/1	8/22	9/1	9/22	10/1	10/22	11/1	11/22	12/1	12/22
1/2	1/23	2/2	2/23	2/23	3/2	3/23	4/2	4/23	5/2	5/23	6/2	6/23	7/2	7/23	8/2	8/23	9/2	9/23	10/2	10/23	11/2	11/23	12/2	12/23
1/3	1/24	2/3	2/24	2/24	3/3	3/24	4/3	4/24	5/3	5/24	6/3	6/24	7/3	7/24	8/3	8/24	9/3	9/24	10/3	10/24	11/3	11/24	12/3	12/24
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1/5	1/26	2/5	2/26	2/26	3/5	3/26	4/5	4/26	5/5	5/26	6/5	6/26	7/5	7/26	8/5	8/26	9/5	9/26	10/5	10/26	11/5	11/26	12/5	12/26
1/6	1/27	2/6	2/27	2/27	3/6	3/27	4/6	4/27	5/6	5/27	6/6	6/27	7/6	7/27	8/6	8/27	9/6	9/27	10/6	10/27	11/6	11/27	12/6	12/27
1/7	1/28	2/7	2/28	2/28	3/7	3/28	4/7	4/28	5/7	5/28	6/7	6/28	7/7	7/28	8/7	8/28	9/7	9/28	10/7	10/28	11/7	11/28	12/7	12/28
1/8	1/29	2/8	3/1	2/29	3/8	3/29	4/8	4/29	5/8	5/29	6/8	6/29	7/8	7/29	8/8	8/29	9/8	9/29	10/8	10/29	11/8	11/29	12/8	12/29
1/9	1/30	2/9	3/2	3/1	3/9	3/30	4/9	4/30	5/9	5/30	6/9	6/30	7/9	7/30	8/9	8/30	9/9	9/30	10/9	10/30	11/9	11/30	12/9	12/30
1/10	1/31	2/10	3/3	3/2	3/10	3/31	4/10	5/1	5/10	5/31	6/10	7/1	7/10	7/31	8/10	8/31	9/10	10/1	10/10	10/31	11/10	12/1	12/10	12/31
1/11	2/1	2/11	3/4	3/3	3/11	4/1	4/11	5/2	5/11	6/1	6/11	7/2	7/11	8/1	8/11	9/1	9/11	10/2	10/11	11/1	11/11	12/2	12/11	1/1
1/12	2/2	2/12	3/5	3/4	3/12	4/2	4/12	5/3	5/12	6/2	6/12	7/3	7/12	8/2	8/12	9/2	9/12	10/3	10/12	11/2	11/12	12/3	12/12	1/2
1/13	2/3	2/13	3/6	3/5	3/13	4/3	4/13	5/4	5/13	6/3	6/13	7/4	7/13	8/3	8/13	9/3	9/13	10/4	10/13	11/3	11/13	12/4	12/13	1/3
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1/19	2/9	2/19	3/12	3/11	3/19	4/9	4/19	5/10	5/19	6/9	6/19	7/10	7/19	8/9	8/19	9/9	9/19	10/10	10/19	11/9	11/19	12/10	12/19	1/9
1/20	2/10	2/20	3/13	3/12	3/20	4/10	4/20	5/11	5/20	6/10	6/20	7/11	7/20	8/10	8/20	9/10	9/20	10/11	10/20	11/10	11/20	12/11	12/20	1/10
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1/24	2/14	2/24	3/17	3/16	3/24	4/14	4/24	5/15	5/24	6/14	6/24	7/15	7/24	8/14	8/24	9/14	9/24	10/15	10/24	11/14	11/24	12/15	12/24	1/14
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1/30	2/20				3/30	4/20	4/30	5/21	5/30	6/20	6/30	7/21	7/30	8/20	8/30	9/20	9/30	10/21	10/30	11/20	11/30	12/21	12/30	1/20
1/31	2/21				3/31	4/21			5/31	6/21			7/31	8/21	8/31	9/21			10/31	11/21			12/31	1/21

Appendix 2

28 Day weaning Schedule

JULY			AUGUST			SEPTEMBER			OCTOBER			NOVEMBER			DECEMBER		
DOB	Wean on (21d)	Wean on (28d)	DOB	Wean on (21d)	Wean on (28d)	DOB	Wean on (21d)	Wean on (28d)	DOB	Wean on (21d)	Wean on (28d)	DOB	Wean on (21d)	Wean on (28d)	DOB	Wean on (21d)	Wean on (28d)
7/1	7/22	7/29	8/1	8/22	8/29	9/1	9/22	9/29	10/1	10/22	10/29	11/1	11/22	11/29	12/1	12/22	12/29
7/2	7/23	7/30	8/2	8/23	8/30	9/2	9/23	9/30	10/2	10/23	10/30	11/2	11/23	11/30	12/2	12/23	12/30
7/3	7/24	7/31	8/3	8/24	8/31	9/3	9/24	10/1	10/3	10/24	10/31	11/3	11/24	12/1	12/3	12/24	12/31
7/4	7/25	8/1	8/4	8/25	9/1	9/4	9/25	10/2	10/4	10/25	11/1	11/4	11/25	12/2	12/4	12/25	1/1
7/5	7/26	8/2	8/5	8/26	9/2	9/5	9/26	10/3	10/5	10/26	11/2	11/5	11/26	12/3	12/5	12/26	1/2
7/6	7/27	8/3	8/6	8/27	9/3	9/6	9/27	10/4	10/6	10/27	11/3	11/6	11/27	12/4	12/6	12/27	1/3
7/7	7/28	8/4	8/7	8/28	9/4	9/7	9/28	10/5	10/7	10/28	11/4	11/7	11/28	12/5	12/7	12/28	1/4
7/8	7/29	8/5	8/8	8/29	9/5	9/8	9/29	10/6	10/8	10/29	11/5	11/8	11/29	12/6	12/8	12/29	1/5
7/9	7/30	8/6	8/9	8/30	9/6	9/9	9/30	10/7	10/9	10/30	11/6	11/9	11/30	12/7	12/9	12/30	1/6
7/10	7/31	8/7	8/10	8/31	9/7	9/10	10/1	10/8	10/10	10/31	11/7	11/10	12/1	12/8	12/10	12/31	1/7
7/11	8/1	8/8	8/11	9/1	9/8	9/11	10/2	10/9	10/11	11/1	11/8	11/11	12/2	12/9	12/11	1/1	1/8
7/12	8/2	8/9	8/12	9/2	9/9	9/12	10/3	10/10	10/12	11/2	11/9	11/12	12/3	12/10	12/12	1/2	1/9
7/13	8/3	8/10	8/13	9/3	9/10	9/13	10/4	10/11	10/13	11/3	11/10	11/13	12/4	12/11	12/13	1/3	1/10
7/14	8/4	8/11	8/14	9/4	9/11	9/14	10/5	10/12	10/14	11/4	11/11	11/14	12/5	12/12	12/14	1/4	1/11
7/15	8/5	8/12	8/15	9/5	9/12	9/15	10/6	10/13	10/15	11/5	11/12	11/15	12/6	12/13	12/15	1/5	1/12
7/16	8/6	8/13	8/16	9/6	9/13	9/16	10/7	10/14	10/16	11/6	11/13	11/16	12/7	12/14	12/16	1/6	1/13
7/17	8/7	8/14	8/17	9/7	9/14	9/17	10/8	10/15	10/17	11/7	11/14	11/17	12/8	12/15	12/17	1/7	1/14
7/18	8/8	8/15	8/18	9/8	9/15	9/18	10/9	10/16	10/18	11/8	11/15	11/18	12/9	12/16	12/18	1/8	1/15
7/19	8/9	8/16	8/19	9/9	9/16	9/19	10/10	10/17	10/19	11/9	11/16	11/19	12/10	12/17	12/19	1/9	1/16
7/20	8/10	8/17	8/20	9/10	9/17	9/20	10/11	10/18	10/20	11/10	11/17	11/20	12/11	12/18	12/20	1/10	1/17
7/21	8/11	8/18	8/21	9/11	9/18	9/21	10/12	10/19	10/21	11/11	11/18	11/21	12/12	12/19	12/21	1/11	1/18
7/22	8/12	8/19	8/22	9/12	9/19	9/22	10/13	10/20	10/22	11/12	11/19	11/22	12/13	12/20	12/22	1/12	1/19
7/23	8/13	8/20	8/23	9/13	9/20	9/23	10/14	10/21	10/23	11/13	11/20	11/23	12/14	12/21	12/23	1/13	1/20
7/24	8/14	8/21	8/24	9/14	9/21	9/24	10/15	10/22	10/24	11/14	11/21	11/24	12/15	12/22	12/24	1/14	1/21
7/25	8/15	8/22	8/25	9/15	9/22	9/25	10/16	10/23	10/25	11/15	11/22	11/25	12/16	12/23	12/25	1/15	1/22
7/26	8/16	8/23	8/26	9/16	9/23	9/26	10/17	10/24	10/26	11/16	11/23	11/26	12/17	12/24	12/26	1/16	1/23
7/27	8/17	8/24	8/27	9/17	9/24	9/27	10/18	10/25	10/27	11/17	11/24	11/27	12/18	12/25	12/27	1/17	1/24
7/28	8/18	8/25	8/28	9/18	9/25	9/28	10/19	10/26	10/28	11/18	11/25	11/28	12/19	12/26	12/28	1/18	1/25
7/29	8/19	8/26	8/29	9/19	9/26	9/29	10/20	10/27	10/29	11/19	11/26	11/29	12/20	12/27	12/29	1/19	1/26
7/30	8/20	8/27	8/30	9/20	9/27	9/30	10/21	10/28	10/30	11/20	11/27	11/30	12/21	12/28	12/30	1/20	1/27
7/31	8/21	8/28	8/31	9/21	9/28				10/31	11/21	11/28				12/31	1/21	1/28

Appendix 3

Spacing Requirements per the Guide for the Care and Use of Laboratory Animals.

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

Animals	Weight, g	Floor Area/Animal,[a] $in.^2$ (cm^2)	Height,[b] in. (cm)	Comments
Mice in groups[c]	<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.[d]
Rats in groups[c]	<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.[d]
Hamsters[c]	<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[c]	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.

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

[a]Singly housed animals and small groups may require more than the applicable multiple of the indicated floor space per animal.
[b]From cage floor to cage top.
[c]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.
[d]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.