

Office of Research Support and Compliance

Vice President for Research, Scholarship and Creative Endeavors

Guidelines for Use of Food or Fluid Restrictions in Mice and Rats

The University of Texas at Austin Institutional Animal Care and Use Committee

These guidelines have been written to assist faculty, staff, and students in performing vertebrate animal procedures in a humane manner and complying with pertinent regulatory requirements. Under some circumstances deviations from these procedures may be indicated but such variances must be approved in advance by the IACUC.

This appendix to IACUC Guideline 21, “Guidelines for the Use of Food or Fluid Restrictions”, provides information to be used when planning and performing fluid regulation in mice and rats used for research, teaching, or testing purposes at the University of Texas at Austin.

Section A – Background Information

Section B – Food/Fluid Regulation Procedures

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Section A – Background

The estimated daily water intake in an adult mouse is 15 ml /100g body weight and in an adult rat is 8-10ml/100g body weight. An adult mouse consumes an average of 12 g/100 g body weight of food per day, whereas an adult rat will consume an average of 5g/100g body weight. Mice and rats consume 70-85% of their food and water during the night cycle, with drinking typically associated with meal consumption (Siegel, 1961). This appendix to IACUC Guideline 21, “Guidelines for the Use of Food or Fluid Restrictions”, provides information to be used when planning and performing fluid regulation in mice and rats used for research, teaching, or testing purposes at the University of Texas at Austin.

Section B – Food/Fluid Regulation Procedures

Prior to introducing Food or Fluid Regulation or manipulation:

1. Investigators must complete the “Procedures for Nonstandard Feeding or Watering” form to communicate restrictions for animals maintained in ARC-managed facilities.
2. Restriction-specific cage cards are available through the ARC and must be fixed to the cage(s) and completed if the ARC is not to provide food or fluid to the animals.
3. After the completed request form is submitted, the protocol will be evaluated by ARC management to verify that nonstandard feeding or watering has been included in the protocol description and approved by the IACUC.
4. The PI or designate will be contacted to arrange for training and implementation of the appropriate daily checklists (as needed) by the ARC supervisor.

Introduction to Regulation:

1. Providing highly palatable fluids (e.g.: sucrose solution, evaporated milk, soy milk) or foods (Fruit loops, Nutella, peanut butter chips) as rewards should be used preferentially over restriction or regulation. Due to neophobia, rodents may need to be introduced to novel foods multiple times to assess preference, as consumption tends to increase over time as neophobia attenuates.
2. Before a restriction/deprivation study is initiated, ad libitum daily food and/or water intake should be determined for that species/strain/sex/age/health status. This information is published for some stocks/strains/ages/etc., of rodents.
3. Animals should be allowed to gradually acclimate to food or fluid restriction (e.g.: 3-5 days transition period).

During Regulation:

1. Daily rations of food and water in an amount and quality to either maintain weight or maintain a suitable growth rate should be a priority.
2. Animals may not be both food and water restricted at the same time.
3. Many experiments involve restricting food or water to motivate animals to perform tasks and are conducted 5 days per week. Therefore, planning for ad libitum access to food and water over the weekend (or days when animals are not working) allows for the recovery of body weight and minimizes health risks associated with the restriction/deprivation schedule.
4. Only trained personnel should monitor and document body weight, body condition, food consumption, hydration status and behavioral changes of the animal. Training on these parameters should be documented on the personnel training records (link)
5. Food and water restriction are prohibited for three days post-surgery, unless specifically justified in the IACUC protocol (to ensure the animal's physiological parameters return to normal before restrictions are introduced).

Section C – Protocol Considerations

1. Scientific justification for the use of restriction or deprivation as well as the planned schedule (Restriction or deprivation schedules described in terms of the duration of restriction/deprivation, time limited access, amount limited access, weekend scheduled access, and/or amount of water provided daily) should be described.
2. Description of measurable parameters used to assess the appropriateness of the level of restriction/deprivation, such as percent ad libitum intake, percent body weight compared to baseline or an unrestricted control animal, or percent body weight compared to an established growth curve.
3. Methods described for assessing and documenting the health and well-being of the animals.
4. If the animals are likely to experience distress resulting from the food and/or water restriction/deprivation procedures, a search for alternative and less stressful procedures must be conducted. Any animal without access to water for over 24 hours must be listed in Category E. Any animal on ongoing fluid regulation with anticipated weight loss of > 20% must be listed in Category E.
5. Criteria to indicate when animals are to be returned to unlimited or increased access to food and/or water.
6. Monitoring schedule for body weights and food/water intake.

Section D – Monitoring/Record keeping requirements

1. Animals must be observed daily, and written records must be maintained for body weight and food and water consumption. Documentation of health assessments must be maintained by the research staff and be easily accessible for review in the housing room.
2. Animals that have restricted access to food or water must be weighed weekly. Twice weekly weighing must be instituted for two weeks after 1) food or water restriction is initiated, 2) food or water restriction is adjusted to decrease the amount of water provided or 4) weight loss is identified when comparing the last two weekly weights.
3. Life stage (e.g., young, growing, pregnant, and lactating) and health status may affect the food and fluid requirement for maintaining rodents. When determining restriction or regulation protocols, these variables should be considered and appropriate monitoring implemented (e.g., weight assessment as well as comparison with expected growth curve or non-restricted controls).
4. Food and water restriction is not recommended in young, growing rodents (e.g., less than 6-8 weeks old) since immature animals are especially sensitive to food and water restriction. If this is scientifically necessary, these animals must be weighed two times per week, and closely monitored and evaluated in relation to normal growth curves and/or unrestricted control animals.
5. Investigators must complete the ARC's Procedures for Nonstandard Feeding or Watering form to communicate restrictions for animals maintained in ARC managed facilities. Restriction specific cage cards are available through the ARC. They must be fixed to the cage(s) and completed daily by the lab if the ARC is not to providing food or water.
6. During active restriction/deprivation periods, records should be available in the animal housing room.

Section E – Criteria for intervention

1. Mice, being one logarithmic unit smaller in body weight than rats, have correspondingly higher basal metabolic rates and correspondingly faster caloric and water turnover. Mice are therefore less tolerant of food and water restriction. Healthy adult mice and rats that have been gradually reduced to 15% of their baseline body weight by restriction/deprivation schedules will rarely show clinical signs of distress. (Rowland, 2007). Weight loss over 20% must be scientifically justified and approved by the IACUC.
2. Animals must be given unrestricted access to fluid if there is >20% body weight loss from baseline (unless approved in the IACUC protocol), the BCS is <2.5/5 (Figure 1), or significant abnormal behaviors have developed.
3. Dehydrated animals (Bekkavold, 2013) require immediate treatment. Supplemental oral fluids or parenteral hydration (such as subcutaneous Lactated Ringer's Solution) must be administered whenever the clinical signs of dehydration are observed.. Contact ARC vetstaff should you have concerns about dehydration in fluid-restricted animals, as parenteral supplemental fluids must be administered gradually to prevent over-hydration.

Physical signs that may be evidence of dehydration include:




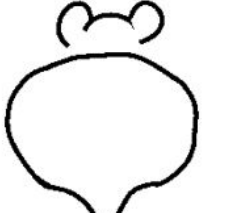

- Anorexia
- scant or no urine output
- scant hard feces
- reduced skin turgor or dry mucous membranes
- behavioral changes such as lethargy or incoordination





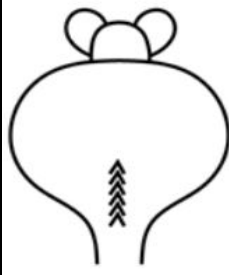
Section F – Food/Fluid Manipulation (Experimental/Non-standard/Special Diets)

The following items must be maintained for each animal on the study:

1. Signage to clearly mark which animal(s) are on a special food/fluid.
2. While on food or fluid manipulation, the ARC will continue to carry out all other husbandry duties including daily health checks and routine cage changing.
3. General information including protocol number, PI, contact person should any issues arise during the provision of the special food/fluid.
4. Health observations (e.g., hydration status, behavior, body condition score, presence of urine and feces, or any other criteria used to determine temporary or permanent removal from the special diet) documented at the frequency described in the protocol (at least once per week).
5. Date, time, and initials of personnel who evaluated animals must be in the animal housing area.
6. If there are anticipated adverse effects, and/or if protocol requirements indicate monitoring adverse effects such as weight loss, then additional monitoring and weighing of the animals may be necessary as described in the protocol or required based on veterinary recommendation.

Body Condition Chart for Laboratory Mice

	BC 1 Mouse is emaciated. <ul style="list-style-type: none">◦ <i>Skeletal structure extremely prominent; little or no flesh cover.</i>◦ <i>Vertebrae distinctly segmented.</i>
	BC 2 Mouse is underconditioned. <ul style="list-style-type: none">◦ <i>Segmentation of vertebral column evident.</i>◦ <i>Dorsal pelvic bones are readily palpable.</i>
	BC 3 Mouse is well-conditioned. <ul style="list-style-type: none">◦ <i>Vertebrae and dorsal pelvis not prominent; palpable with slight pressure.</i>
	BC 4 Mouse is overconditioned. <ul style="list-style-type: none">◦ <i>Spine is a continuous column.</i>◦ <i>Vertebrae palpable only with firm pressure.</i>
	BC 5 Mouse is obese. <ul style="list-style-type: none">◦ <i>Mouse is smooth and bulky.</i>◦ <i>Bone structure disappears under flesh and subcutaneous fat.</i>
<p>A "+" or a "-" can be added to the body condition score if additional increments are necessary (i.e. ...2+, 2, 2-...)</p>	

	<p>BC 1</p> <p>Rat is emaciated</p> <ul style="list-style-type: none">• Segmentation of vertebral column prominent if not visible.• Little or no flesh cover over dorsal pelvis. Pins prominent if not visible.• Segmentation of caudal vertebrae prominent.
	<p>BC 2</p> <p>Rat is under conditioned</p> <ul style="list-style-type: none">• Segmentation of vertebral column prominent.• Thin flesh cover over dorsal pelvis, little subcutaneous fat. Pins easily palpable.• Thin flesh cover over caudal vertebrae, segmentation palpable with slight pressure.
	<p>BC 3</p> <p>Rat is well-conditioned</p> <ul style="list-style-type: none">• Segmentation of vertebral column easily palpable.• Moderate subcutaneous fat store over pelvis. Pins easily palpable with slight pressure.• Moderate fat store around tail base, caudal vertebrae may be palpable but not segmented.
	<p>BC 4</p> <p>Rat is overconditioned</p> <ul style="list-style-type: none">• Segmentation of vertebral column palpable with slight pressure.• Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis palpable with firm pressure.• Thick fat store over tail base, caudal vertebrae not palpable.
	<p>BC 5</p> <p>Rat is obese</p> <ul style="list-style-type: none">• Segmentation of vertebral column palpable with firm pressure; may be a continuous column.• Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis not palpable with firm pressure.• Thick fat store over tail base, caudal vertebrae not palpable.

Section G – References

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