

Food and Fluid Regulation

Policy: This policy establishes standards and expectations for researchers when employing food and/or fluid regulation for experimental purposes. The regulation of food and/or fluid may be required for some physiological, neuroscience, and behavioral research studies. The regulation process may involve *scheduled access* to food or fluid sources, or *restriction* of the total volume of food or fluid consumed. All studies employing food and/or fluid regulation must provide scientific justification for the necessity *and* duration of the regulation within the *IACUC Proposal*. The least regulation necessary to achieve scientific aims should be utilized. Animals undergoing regulation must be closely monitored to ensure that food and fluid intake meets their nutritional needs. This policy **does not** apply to “periods of fasting of 24 hours or less, preceded or proceeded by *ad libitum* access to palatable food and fluid sources of at least forty-eight hours”² or to animals restricted/regulated for the maintenance of healthy body weight, however such details must still be included in the *IACUC Proposal*. This policy **does not** pertain to animals restricted/regulated under the direction of RRF veterinary staff or to standard pre-anesthetic fasting.

Rationale: The *Guide for the Care and Use of Laboratory Animals* (Guide) recognizes that food and/or fluid restriction or regulation may be necessary for certain experiments. The Guide states that these studies should “use the least restriction necessary to achieve the scientific objective while maintaining animal well-being.” The Animal Welfare Act and Regulations allow for short-term withholding of food or water “when specified in an IACUC-approved activity that includes a description of monitoring procedures.”³ The IACUC is responsible for reviewing and approving all food and/or fluid regulation prior to implementation. This policy has been developed to serve as a resource for researchers, animal caregivers, and IACUC members in determining how to appropriately conduct food and fluid regulation in a manner consistent with animal welfare and regulations, while not compromising data collection.

Procedures, Guidelines, and Exceptions:

1. All food or fluid regulation must be detailed in the *IACUC Proposal*, and reviewed and approved by the IACUC prior to implementation. The least amount of restriction necessary to achieve study objectives must be used.
2. USDA-regulated species may not be both food and fluid restricted.
3. *Proposals* requesting food and/or fluid regulation should address:
 - a. The type and duration of regulation to be employed.
 - b. Thorough justification detailing the necessity and the duration of regulation. The justification should establish why the regulation is necessary to accomplish research objectives and include an assurance that it will occur for the shortest time possible.
 - c. Details regarding plans for monitoring, weighing, and assuring adequate nutrition and hydration.
 - d. Documented consideration of alternatives to food and/or fluid regulation.
 - e. Criteria must be defined for temporary or permanent removal of animals from the study.
4. Scheduled regulation should make food and/or water available for at least 15 minutes a day.

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5. Animals undergoing surgical procedures must receive *ad libitum* food/water at least one week prior to and following the surgical procedure. However, pre-anesthetic fasting or water restriction may be acceptable. Rabbits and rodents are not typically fasted prior to anesthesia.
6. In the case of conditioned-response research *Proposals*, use of a highly preferred food or fluid as a positive reinforcement, instead of restrictions, is recommended. ¹
7. Each cage or run must be marked to indicate that the animal is under food or fluid regulation. A start time and end time for fasting or time of scheduled feeding(s) should be clearly identified. For rodents, a green “dietary alert” card with this information should be placed on the cage.

Special Considerations for Rodents:

1. Rodents should be acclimated over at least three days to the new regulation/scheduling; food and/or fluid should be gradually reduced over the acclimation period.
2. Regulation is not recommended in rodents under 12 weeks of age and requires additional scientific justification.
3. Rodents cannot be completely deprived of food or water for more than 24 hours.
4. Food and water must be made available concurrently, as rodents typically do not eat without available water.

Food Regulation:

1. Animal can be fed 70% of *ad libitum* food consumption until they reach 85% baseline weight. Restriction down to 50% of *ad libitum* food consumption is not recommended, but may be scientifically justified in certain situations.
2. *Ad libitum* food consumption can be established by weighing the food daily or using published standards of animals matching strain, sex, and age.
3. Special attention should be given to ensure that intake meets the animal’s nutritional needs to maintain well-being.

Fluid Regulation:

1. Fluid regulation can vary widely based on the species and task. Restricted fluid regulation usually involves permitting a percentage of *ad libitum* fluid intake be consumed outside of the testing period.
2. The total fluid consumed within and outside the testing period must be sufficient to maintain well-being.
3. Animal observed as clinically dehydrated (listless, inactive, increased skin tent, and/or sunken eyes) must be immediately offered drinkable fluid support along with an alternative fluid support such as subcutaneous fluids, moist chow, or Napa Nectar™ gel. Additionally, the animal should be reported to RRF veterinary staff.

Monitoring & Humane Endpoints:

1. Baseline body weight must be measured and recorded *before* food or water regulation occurs. Body weight must be recorded at least weekly and more often for animals requiring greater restriction. Daily body weight measurements are recommended for rodents.
2. A monitoring program must be established for each animal that includes daily observation during regulation (including weekends and holidays). Lab members involved in fluid regulation should be trained and able to identify signs of dehydration.

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3. If an animal appears dehydrated, listless, hunched, or is showing signs of pain/distress, RRF veterinary staff must be contacted immediately in addition to providing supplemental food/water.
4. Upon reaching 85% of a baseline weight, animals should no longer be heavily restricted.
5. If the goal is to maintain animals at less than or equal to 85% of expected weight, Pain and Distress Class E (Formerly Class III) categorization is recommended and additional justification is required.
6. No food or water regulation should occur once reaching 80% of baseline weight.

Recordkeeping:

Labs must maintain a written record for each animal undergoing food or fluid regulation. The records must be made available to the IACUC and RRF staff and must include:

- a. The baseline weight of the animal before regulation
- b. Date (record must be updated daily during regulation period)
- c. Daily food consumption (g)
- d. Daily water consumption (ml)
- e. Weekly weight (or more often for animals requiring greater restriction)
- f. Daily health observations (e.g., hydration status, behavior, body condition score, or any other removal criteria identified in the *Proposal*)
- g. Initials of observer/recorder

References:

1. National Research Council, Guide for the Care and Use of Laboratory Animals, 8th Ed., National Academies Press, Revised 2011.
2. National Institutes of Health Office of Intramural Research (Office of Animal Care and Use), Guidelines for Diet Control in Laboratory Animals, Revised 2019.
3. USDA Animal and Plant Health Inspection Service, Animal Welfare Regulations. 9 CFR Part 2.38(f)(2)(ii), 2016.

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