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IACUC GUIDELINE:	FOOD AND WATER REGULATION
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Background:

Animal studies may occasionally require the control of food or fluid intake to achieve a specific experimental goal, for example, studies of homeostatic regulation of energy metabolism or fluid balance, studies of the motivated behaviors and physiological mediators of hunger or thirst, and studies that control food or fluid consumption to motivate animals to perform novel or learned tasks. However, any use of food or fluid control to achieve a scientific objective must comply with regulatory standards:

The *Guide for the Care and Use of Laboratory Animals*, 8th Edition (2011), states:

"Animals should be fed palatable, uncontaminated diets that meet their nutritional and behavioral needs at least daily, or according to their particular requirements, unless the protocol in which they are being used requires otherwise. Subcommittees of the National Research Council Committee on Animal Nutrition have prepared comprehensive reports of the nutrient requirements of laboratory animals (NRC 1977, 1982, 1993, 1994, 1995a, 1998b, 2000, 2001, 2003a, 2006b,c, 2007); these publications consider issues of quality assurance, freedom from chemical or microbial contaminants and natural toxicants in feedstuffs, bioavailability of nutrients in feeds, and palatability."

"Animals should have access to potable, uncontaminated drinking water according to their particular requirements."

"The development of animal protocols that involve the use of food or water regulation requires the evaluation of three factors: the necessary level of regulation, potential adverse consequences of regulation, and methods for assessing the health and well-being of the animals…"

"The animals should be closely monitored to ensure that food and fluid intake meets their nutritional needs… Written records should be maintained for each animal to document daily food and fluid consumption, hydration status, and any behavioral and clinical changes used as criteria for temporary or permanent removal of an animal from a protocol. In the case of conditioned-response research protocols, use of highly preferred food or fluid as positive reinforcement, instead of restriction, is recommended."

The USDA Animal Welfare Act [9 CFR, Subchapter A, Part 2, Section 2.38 (f)(ii)] states:

"Deprivation of food or water shall not be used to train, work, or otherwise handle animals; Provided however: That the short-term withholding of food or water from animals, when specified in an IACUC-approved activity that includes a description of monitoring procedures, is allowed by these regulations."

Definitions:

- Regulation is defined as offering the animal special diets or treated water (i.e., something other than ad libitum standard rodent chow and untreated drinking water) for experimental reasons.
- Restriction is defined as offering less than ad libitum access to food or water and less than the normal daily food or water intake for the species.
- Deprivation is defined as withholding food or water for greater than 24 hours.

IACUC Expectations:

To receive IACUC approval for food or water regulation, restriction, or deprivation, investigators must address the following subjects in their IACUC protocol application:

1. A scientific justification for the food/water regulation, deprivation, and/or restriction.

2. A description of how the food and/or water regulation, restriction or deprivation will be accomplished, including due consideration for:
    - Assurance that a palatable, uncontaminated diet that meets the nutritional requirements of the animals is being offered.
    - Assurance that a non-palatable water treatment is not toxic or causes adverse effects.
    - The least restriction possible to achieve the scientific objectives will be used.
    - The use of highly palatable or preferred food or fluid as a positive reinforcement will be used instead of restriction, if possible.

3. A description of the monitoring procedures that will be used to assess the health and well-being of the animals and prevent them from becoming dehydrated and/or malnourished, including provisions for:

- Daily monitoring is recommended.
- The routine weighing of all animals, which should be not less than once weekly.
- Establishing target weights or growth rates that are species-, age-, sex-, and strain-specific. However, target weights or growth rates need not be derived from comparisons to animals with ad libitum access to food, since in most cases ad libitum access is not optimal for long-term health.[2, 3] A sensible target growth rate for adult (between 20 and 113 weeks of age) Sprague Dawley rats on food restrictions might be 2 g/wk in males, and 1 g/wk in females.[3] A commonly used target body weight for mice undergoing water restriction for neuroscience experiments is 85% of reference weight.[5] Reference weights for mice should be established from the free-feeding weight of the animal before starting fluid control. When the reference weight is established, these mice should be greater than 8 weeks of age (i.e., not still growing). Reference weights should be periodically updated during breaks from the water restriction longer than 7-10 days when the mice are offered ad libitum water.[5]
- The criteria (preferably objective and well-defined) for the temporary or permanent removal of animals from deprivation and/or restriction conditions. For mice undergoing water restriction for neuroscience experiments, these criteria commonly include body weight below 80% of the reference value for several days, reduced activity in the home cage, and skin tenting (indicated severe dehydration).[5]

References:

1. *Guide for the Care and Use of Laboratory Animals*, 8th Edition. National Academies Press. 2011. Pages 31, 65, and 68.

2. Keenan KP, Hoe CM, Mixson L, McCoy CL, Coleman JB, Mattson BA, Ballam GC, Gumprecht LA, and Soper KA. 2005. Diabesity: a polygenic model of diatery-induced obesity from ad-libitum over-feeding of Sprague-Dawley rats and its modulation by moderate and marked dietary restriction. Toxicol Pathol 33:650-674.

3. Rowland NE. 2007. Food or fluid restriction in common laboratory animals: balancing welfare considerations with scientific inquiry. Comp Med 57(2):149-160.

4. Goldstein PM, Reinert S, Glas A, Bonhoeffer T, and Hübener M. 2018. Food and water restriction lead to differential learning behaviors in head-fixed two-choice visual discrimination task for mice. PLOS One. https://doi.org/10.1371/journal.pone.0204066

5. Barkus, C, et. al. 2022. Refinements to rodent head fixation and fluid/food control for neuroscience. J Neurosci Methods. https://doi.org/10.1016/j.jneumeth.2022.109705