

Policy on Intake Regulation Guidelines for Rats and Pigeons

Categorization of Protocols

Food and/or water restriction studies conducted in **healthy** rats or pigeons will be considered category 1 studies if 1) the floor weight is not less than 80% of the body weight from established growth curves, unrestricted control animals or, in the case of adults, pre-study control weights 2) the target weight is not less than 85% of this established body weight and 3) the experimental treatment is not expected to produce further weight loss than the restriction alone. Deviation from these conditions will rank the protocol as category 3.

Food and/or water restriction studies in other species will be considered category 3 unless the IACUC determines otherwise.

Background:

1. Food or fluid restriction may be required in order to achieve a variety of research and/or husbandry objectives.
 - a. Behavioral research protocols may require withholding food and/or water either in order to train animals to perform a task while providing food or water as a reward for correct behavior or because food or fluid intake after a fast is the behavior under investigation.
 - b. Nutritional studies may require altering the levels of specific nutrients.
 - c. Limiting feeding is common for some sedentary laboratory species in order to control obesity.
 - d. Food may be withheld for a specified period prior to surgery in order to prevent vomiting and aspiration of food during or after anesthesia.
 - e. Restricted feeding under “pair-feeding” and related feeding conditions in order to differentiate the expected effects of an experimental treatment from anorexia as a side effect.
2. It has been shown that some methods of food and/or fluid restriction may be physiologically and/or psychologically stressful. If restriction is allowed to exceed acceptable levels it can be physically harmful to an animal.

Purpose:

These guidelines were developed to provide general guidance for investigators and IACUC members. Food and/or fluid restriction studies involving other laboratory animal species must be considered on a case-by-case basis.

The goal of these guidelines is to ensure that:

- a. Dietary restrictions are well justified and necessary for study objectives.
- b. Animals subjected to dietary restriction are appropriately monitored.

Definition:

For the purpose of these guidelines, food or fluid restriction will include any deviation from the normal husbandry or other continuous access procedures.

Guidelines:

1. Food and/or fluid restriction that deviates from the normal husbandry procedures must be described in the Animal Care and Use Protocol (ACUP) and approved by the IACUC.
2. In non-rodent species, food and/or water deprivation for a period of greater than 24 hours requires scientific justification, a literature search for alternatives and is considered a pain/distress category 3 (USDA – E). While the definition of pain is clearly defined, the definition of distress for the purpose of this policy is, “A state in which an animal cannot escape from or adapt to the external stressors or conditions that it experiences resulting in negative effects on its well-being” (NRC Workshop, 2000).
3. The rationale for restricting food or water intake may be to establish food or water as positive reinforcement, to study caloric restriction, or to prevent obesity and protect the health of the animals. Restrictions must be conducted with care and tailored to the feeding patterns and nutritional requirements of the strain and species as well as the requirements of the study.

Rodents and many other species adapt well to once per day feeding. Daily rations in an amount and quality to either maintain weight at a specified minimum (see below) or to maintain a suitable growth rate should not be viewed as restriction of intake.

4.
 - a. In conditioned response research, use of highly preferred food as positive reinforcement can be used instead of food restriction.
 - b. Whenever an animal obtains any portion of its diet through positive reinforcement (food reward), the investigator must ensure that the sum of the nutritional value of the food earned through reward and of the food provided “free” (without necessity of earning it) is sufficient to maintain the animal in a healthy state. Consideration should be given to the use of a food reward which is sufficiently desirable and motivating for the animal

- that dietary restriction is unnecessary. However, dietary restriction may be justified in some cases; depending on the species, the behavioral task, and the requirements of the research protocol.
5.
 - a. Water deprived animals often have non-restricted access to food, but investigators should be aware that most food consumption of dry food declines progressively with time since water was last available. Water should be available for long enough to maintain sufficient food intake.
 - b. Water restriction should be avoided unless scientifically justified.
 6. The investigator is responsible for assuring that specially formulated diets are nutritionally adequate and palatable.
 7. The investigator must monitor parameters such as body weight, hydration status, body condition, and food consumption with suitable frequency.
 8.
 - a. The investigator must indicate in the protocol the percentage changes to arrive at the target and floor weights in the protocol. The floor weight is defined as the weight below which the investigator must immediately notify the ACS veterinary staff and provide supplementary feeding of the animals.
 - b. Intervention endpoints must be specified in the protocol. Examples of specific endpoints include:
 - i. Failure of animals that would normally be growing to gain weight.
 - ii. Loss of greater than 15% of the body weight of age- matched animals as determined by comparison with established growth curves or growth rates of unrestricted control animals. It should be specified when the animals will be weighed in relation to when they are fed.
 - iii. For mice and rats, a body condition score of 2 is underconditioned:
 - (a) segmentation of vertebral column is evident;
 - (b) dorsal pelvic bones are readily palpable. ([Ref: Lab Animal Science, 1999; 49\(3\): 319-323](#)); Uliman-Cullere MH, Foltz CJ)
 9. The investigator must specify that calibrated scales will be used, the scale is validated with a known weight to ensure accuracy at a monthly minimum, and calibration records are maintained for inspection purposes.
 10. Weight records of all animals on food/water restriction must be maintained in animal housing rooms.

Specific Requirements:

1. Conditioned Response Research Protocols

- a. The amount of food/fluid restriction used must be the minimum level that will achieve the objective.
- b. If food/fluid restriction is to be used in an experimental protocol, the method of restriction and scientific justification for its use must be clearly explained in the Animal Care and Use Protocol submitted for review and approval by the IACUC,
- c. Restriction must be based on a measurable parameter such as percentage of *ad libitum* intake, percentage of body weight compared to an unrestricted control animal, percentage of body weight compared to an established growth curve, or length of time access to food/fluid is allowed per 24 hours.
- d. Consideration must be given to alternative methods and/or modifications to food and/or fluid restriction.
- e. In order to make a knowledgeable determination of an appropriate level of food/fluid restriction, it is necessary to know what normal quantities of food or fluid are required for maintenance of the species. Life stage (growth, pregnancy, lactation, and senescence), demands of the task, and state of health must also be taken into consideration in determining maintenance requirements.
- f. Unless scientifically justified, food and/or water restriction leading to a body weight loss of greater than 10% must be introduced incrementally to allow for physiological and psychological adaptation.

2. Nutrition Studies:

- a. The principal investigator is responsible for ensuring the proper formulation and nutritional adequacy of these diets.
- b. Specific arrangements for feeding and diet storage must be provided in the ACUP and arranged with Animal Care Services.
- c. These rations frequently vary in form and in palatability. The animals must be closely monitored to insure that an adequate amount is consumed.

3. Pre-anesthetic Fasting:

- a. Pre-anesthetic fasting is not required for rodents and rabbits. However, specific surgical procedures or experimental designs that require an overnight fast must be justified in the ACUP.
- b. For non-rodent and non-rabbit species, food may be withheld up to 24 hours prior to an anesthetic procedure (i.e. overnight fasting). Water must be available during the overnight fast but may be removed in the morning on the day of surgery.

4. Restricted Feeding to Control Obesity:

- a. The quantity fed should be appropriate for maintaining the weight of mature adult animals.
- b. As a program to restricted-feeding is established, more frequent monitoring of animal weights will be required to determine an appropriate amount of feed; once the program is established, less frequent monitoring is acceptable.

5. Dietary Restriction for Other Reasons:

- a. An explanation of the need for diet or fluid restriction must be provided in the ACUP along with a description of the nature of the restriction.
- b. Consideration must be given to alternative methods and/or modifications to food and/or fluid restriction.

6. Documentation must be provided:

- a. Document in the ACUP whether animals are weighed pre- or post-feeding.
- b. Weight records are to be maintained and monitored by the PI or the PI's designee.
- c. Weight records should be available in the animal housing room for monitoring by the animal care staff.

References:

NIH Methods of Welfare Consideration in Behavioral Research with Animals
Keenan, K.P., Ballam, G.C., Haught, D.G. and Laroque, P. (2000) Nutrition. In G.J. Krinke (Ed.) *The Laboratory Rat*. N.Y.: Academic Press.