

University of Maryland College Park Animal Care and Use Policy

# **Guidelines for Food and Water Restriction**

### Introduction

The purpose of these guidelines is to assist investigators in the development of procedures when experimental methodology requires the restriction of food or water.

### Considerations

- In the development of protocols utilizing food or water restriction, investigators should address the necessary level of restriction, the potentially adverse consequences of restriction (level of pain and distress) and the methods for assessing the health and well-being of the animals.
- Consideration must be given for the species, strain and the animal's size, age, health status, body condition, and hydration status.
- Any additional dietary requirements of young, developing animals when maintaining their normal rate of growth must be considered.

### Procedures – General

- The details of the paradigm used and accountability of the individuals involved must be clearly outlined in the approved animal study protocol.
- Clear justification is required in the animal study protocol if any period of 12 hours or more of reduced food or fluid intake is required. Justification is also required for the extent and duration of the restriction.
- Withholding food and fluids prior to anesthesia from small animals such as rodents may not be necessary because rodents are incapable of vomiting. Additionally, rodents require continuous access to energy.
- Withholding food and fluids prior to anesthesia in other animals is dependent on the species. Ferrets only require a 2 hour withdrawal for food and water while sheep may require food to be withheld for 36 hours and fluids for 18 hours.
- Animals on feed control should be allowed a short-term unrestricted feeding period prior to any anesthetic procedure to avoid the development of hypoglycemia during the recovery period.
- A plan of action to maintain the health and well-being of the animals, complete with endpoints for therapeutic intervention, should be established. This may include "pull" weights where, when reached, the animal is given supplemental food or pulled from the study.
- Investigators, animal care staff and veterinarians working with animals on

food or water controlled access paradigms must know the species-typical signs of distress for the animals with which they are working.

- Animals routinely adapt well to the research design and display few signs of distress. However, animals must be carefully monitored on a daily basis to ensure that they are healthy, adapting normally, and consume sufficient food and/or water to maintain good health.

## **Procedures - Food**

Food Reinforcement

- Whenever an animal obtains any portion of its diet through food reinforcement, the

sum of the nutritional value of the food earned through reinforcement and of the food provided "free" (without the necessity of earning it) must be determined to be sufficient to maintain the animal in a healthy state.

- When possible, the food reinforcement should be a substance and size that is sufficiently reinforcing and motivating (e.g., raisins, peanuts) such that dietary restriction is minimized or unnecessary.

Food Restriction

- In most cases, some food should be provided every day unless a specific exception to this policy has been obtained in an approved animal study protocol.
- Experience has demonstrated little adverse consequences to a short period (24 hours) without food intake in normal healthy animals.
- Experience has demonstrated that short periods, generally 48 hours or less, of markedly reduced food intake or fasting may be required during the initial phases of diet control, or after periods of increased food intake (e.g., ad libitum food availability).

- When caloric control is an experimental requirement, other aspects of the animals' diet should remain balanced (e.g., vitamins, minerals, etc.).

- Weight records must be kept for animals on dietary control, a minimum of once each week. If an animal shows a loss in body weight of more than 15 to 20% during the period of study, when compared to the pre-diet control weight of the animal, the animal must be evaluated by the attending or facility veterinarian and, if deemed necessary, its food increased appropriately. Exceptions to this policy are allowed only if the veterinarian determines that the weight loss does not endanger the animal's health. One example of an exception in an obese animal that is placed on caloric restriction. When evaluating an animal with a weight loss that was previously obese, the veterinarian may determine a weight of the animal that is closer to its "ideal" weight for the animal.
- An animal's weight should be gradually reduced to a target weight and acclimated to the

feeding schedule to mitigate a stress response. Ideally, the diet restriction should be limited so that the body weight is reduced not more than 5 to 10% per week. Special attention should be given to ensure that the diet fed meets the animal's nutritional needs. In general, the total caloric intake of a food-regulated animal is 50-70% of that associated with ad libitum feeding.

- Physical evaluation of the animal by a veterinarian, changes in palpable muscle mass and evaluation of serum chemistry (e.g., serum protein, albumin levels, etc.) can be helpful for assessing clinical health in animals under dietary control. In addition, it may at times be helpful to monitor an animal for signs of ketosis or metabolic acidosis.
- It may be advisable in some long-term research designs involving diet control to intermittently allow animals a period of ad libitum feeding sufficient to establish a new unrestricted feeding body weight plateau. This may be necessary if the animal stops performing or the veterinarian determines that the animal's current weight endangers its health. When transitioning an animal from a controlled food access paradigm to ad libitum access, careful monitoring of the animal's dietary intake is recommended to aid in the prevention of deleterious gastrointestinal complications. If animals are subsequently placed on diet restriction, the new unrestricted feeding weight may be less than the previous one, and a physical and clinical exam could be warranted.

#### **Procedures – Fluid Restriction**

- At the start of a new research protocol, the body weight should be recorded daily for each animal.
- The transition of an animal to a controlled water access paradigm is best accomplished through a gradual, systematic limitation of fluid intake over a several-day period.
- Whenever an animal obtains any portion of its fluid requirements through fluid reinforcers, the sum of the fluid earned through reinforcement and the supplemental fluid provided outside of the experiment must be is sufficient to maintain the animal in a healthy state.
- When possible, concurrent with the systematic limitation of available free-choice water, animals should be provided with an opportunity to work for additional water until satiated.
- Restriction often may be relaxed or reduced after the animal becomes proficient at a given task.
- Some animals on controlled fluid paradigms are provided with "vacations". A "vacation" is a period of time, ranging from a day to a few weeks in duration, when the animal is provided a markedly increased fluid allocation, commonly >1.5-3 times their routine daily consumption. Following a "vacation" period, an animal may require a period without fluid intake to regain the motivation to perform its learned task.
- When an animal is not required to perform its learned task for prolonged periods of time, (several weeks duration or longer), gradually increasing the animal's consumption to ad libitum access maybe recommended. It is also recommended that animals be provided additional access to fluid on days when research procedures are not scheduled, unless justifiable reasons preclude such fluid supplementation.
- Short periods with or without markedly reduced fluid intake may be required during the initial phases of a research design requiring water control. The duration of the period will vary with the species and hydration status of the animal.
- Experience has demonstrated little adverse consequences to a short period (12 hours) without water intake in normal healthy animals.
- Animals should be acclimated to easy tasks prior to their systematic and gradual progression to more difficult tasks. Once a baseline fluid intake has been established on a given task, each animal should be allowed to earn fluids to satiety or its fluid intake should be appropriately supplemented on a daily basis. In cases in which supplements are required, the minimum amount of fluids to be provided each day should be equivalent to the amount typically consumed by the animal when it is permitted to earn fluids to satiety.
- To ensure the animal's welfare and experimental integrity, daily adjustments in fluid intake may be required during the course of the research.

- Once an animal has learned a behavior, the daily amount of fluid provided should be increased to the maximum level that will ensure adequate and reliable performance of the task.

- Daily records of fluid intake must be maintained and be available for review by the veterinary staff and the IACUC. The daily record should indicate the fluid earned during the recording session and any supplemental fluid and/or fruit provided to the animal.
- Each animal under fluid control must be observed daily for its health status by the animal care or investigative staff.
- Normal physiological responses to fluid control routinely result in changes in the animal's clinical pathological status. For example, fluid control will often result in

elevated blood parameters (e.g., hematocrit, serum total protein, etc.), while physical and behavioral assessment of the animal indicates that the animal is healthy and adapting normally to the controlled access paradigm. If at any time the facility or attending veterinarian determines that an animal is not adapting sufficiently to the controlled fluid paradigm, the veterinarian will consult with the investigator to develop a plan to maintain the health of the animal.

Some animals on a controlled fluid access paradigm may decrease their total caloric intake in response to changes in their access to water. Because food intake is correlated to the amount of fluid consumed, monitoring food consumption can also be a valuable tool. In some cases, the decreased caloric intake is minor and does not result in a body weight loss greater than 15 to 20%. This weight loss may not pose a problem in the case of obese individuals, but could lead to complications in the case of a chronic fluid deficiency. Therefore, as a precaution against chronic fluid deficiency, the animal's weight must be measured and recorded at no less than weekly intervals. If an animal shows a loss in body weight of more than 15 to 20% during the period of study, when compared to the pre-diet control weight of the animal, the animal must be evaluated by a veterinarian and, if required, its fluids or food increased appropriately. When evaluating a previously obese animal with a weight loss, the veterinarian may determine a weight of the animal that is closer to its "ideal" weight for the animal.