STANDARD OPERATING PROCEDURE #408 WATER CONTROL IN NON-HUMAN PRIMATES MACAQUES

1. PURPOSE

This Standard Operating Procedure (SOP) describes the implementation and withdrawal of water control in nonhuman primates (NHPs) specifically macaques (Macaca mulatta and Macaca fascicularis).

2. RESPONSIBILITY

Principal investigator (PI), and their research staff, veterinarian,

Establishment of water consumption baseline:

- 4.6.1. The establishment of the water consumption baseline should be performed under the oversight of the veterinary care staff.
- 4.6.2. When animals are switched from an automatic watering system to drinking from water bottle, allow for a minimum acclimation period of 7 consecutive days.
- 4.6.3. A period of at least 7 consecutive days is required to establish an *ad libitum* baseline for water consumption and the baseline bodyweight.
- 4.6.4. Experimental manipulations should be avoided during this period.

Implementation of water control:

- 4.7.1. After a stable *ad libitum* baseline is established, water control may commence.
- 4.7.2. The transition from unrestricted to controlled fluid access must be accomplished gradually, over a period of several days.
- 4.7.3. Body condition, weight, and hydration status must be assessed daily by the veterinary care and research staff.
- 4.7.4. Starting with the *ad libitum* baseline, the total volume of water consumed can be gradually reduced until the desired performance is achieved.

The total volume of available water can continue to be decreased if the animal does not exhibit weight loss exceeding 15% of its baseline body weight or any clinical signs of dehydration as described in section 4.10.

Fluid should not be reduced to less than 20 ml/kg/day.

If the animal does not consume this amount during testing, supplemental water must be given to meet the minimal total volume.

- 4.7.5. When problems in training or maintaining a behavior are encountered, all variables, such as equipment malfunctions, programming errors, illness, poor animal training skills, or inadequate task criteria for the animal, should be considered before further reducing the volume of water provided. Excessive water deprivation can be detrimental to task performance. Consult the veterinarian before proceeding.
- 4.7.6. Once the animal is fully trained to perform the task, it will be allowed to work for as much water as it wants every working day. This is the new *testing* baseline.
- 4.7.7.

- 4.9.2. Gradually increase the amount of water available over a period of several days or weeks.
- 4.9.3. Starting from the *testing* baseline, increase water availability daily by 20 ml/kg increments until the animal is receiving its *ad libitum* baseline.
- 4.9.4. After reaching the ad libitum baseline, the animal can then have free access to unlimited water.

Ongoing health monitoring:

- 4.10.1. Chronic water control results in the establishment of new physiological set points. Most animals experience an initial weight loss, usually not exceeding 15% of baseline body weight, followed by weight gain and eventual stabilization of body weight.
- 4.10.2. The following parameters must be monitored as specified below:

Body weight:

- 4.10.2.3.1. Evaluated at least once weekly.
- 4.10.2.3.2. Must be obtained in as consistent a manner as possible (e.g. on awake animals, at the same time each day, in a chair with a known weight) each time the animal is removed from its cage.
- 4.10.2.3.3. Each time an animal is anaesthetized, body weight must be recorded.
- 4.10.2.3.4. Body weights can vary by as much as 1kg, depending on whether the animal was fed or watered. Daily weighing must take place at approximately the same time each day in order to obtain consistent and accurate results.
- 4.10.2.3.5. Consult the veterinarian if weight loss exceeds 10%.

Body condition:

- 4.10.2.4.1. Once weekly, veterinary care staff must perform a cage side assessment (e.g., thin, normal, overweight).
- 4.10.2.4.2. Body condition scoring may be performed when the animal is anesthetized.

Hydration status:

- 4.10.2.5.1. Once daily, animal care or veterinary care staff must perform cage side assessment through qualitative observation of fresh feces, urine output, and mental status.
- 4.10.2.5.2. Where there is indication of possible dehydration, the hydration status should be verified through blood osmolality, which should be <320 mOsmol/kg H₂O.

Behavior:

4.10.2.6.1. Observations should be made of any change in behavior, anorexia, depressive behavior, urine drinking, etc.

Hematology and chemistry profiles:

4.10.2.7.1. As determined by the veterinarian, with special attention paid to hematocrit, proteins, renal parameters, and electrolytes.

Additional considerations:

4.11.1. During the first 4 to 6 years of a monkey's life, growth spurts may lead to weight gain of as much as 2kg in a 12-month

SOP REVISION HISTORY

DATE	NEW VERSION
2019.03.27	4.3.2. The establishment of the water consumption baseline should be performed under the oversight of the veterinary cannot be stables as a second of the water consumption baseline should be performed under the oversight of the veterinary cannot be setables.
2019.03.27	4.4.8.On days when animals are not required to work (e.g., weekends), an amount of water equivalent destinated libitum baseline will can be provided. Water control is restarted one day before testing resumes.
2019.03.27	4.4.9. Pair-housed or grouphoused animals should be separated during the day to control/monitor water consumption. They can remair-housed or grouphoused overnight without water.
2019.03.27	4.6.2.1.1. Evaluated at least once weekly.
2019.06.17	3.2. Water is used as a reward during operant conditionthingst becomes a motivator for performance.

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