SAN DIEGO STATE UNIVERSITY

Office of Laboratory Animal Care

IACUC Guidelines

Assessing the Health and Condition of Laboratory Rodents

Date Implemented: 2/12/2024
Date(s) Revised:

PURPOSE/BACKGROUND: Good science and good animal care go hand in hand. A sick or distressed animal does not produce the reliable experimental results that a healthy and unstressed animal produces. Understanding how to assess the health and well-being of animals used in research ensures standards of quality in animal healthcare, is critical for effective communication between Veterinary, Office of Laboratory Animal Care (OLAC), and Laboratory staff, and increases the uniformity and usefulness of experimental data gathered. Understanding how to assess the health and condition of laboratory rodents, as well as how to describe their condition, is also critical to detailing endpoint criteria and evaluation. Detailed endpoint criteria are federally mandated and must be included in approved IACUC protocols.

RESPONSIBLE INDIVIDUALS
It is the responsibility of all those working with animals in research to ensure their health and well-being. In various circumstances both Laboratory and OLAC Staff may be required to assess the health and well-being of mice that are used in research. This level of responsibility and care is mandated by the Public Health Service based on the ***Guide for the Care and Use of Laboratory Animals*** *(National Research Council. 2011).* According to the ***Guide****, "Studies that may result in severe or chronic pain or significant alterations in the animals' ability to maintain normal physiology, or adequately respond to stressors, should include descriptions of appropriate humane endpoints or provide science-based justification for not using a particular, commonly accepted humane endpoint. Veterinary consultation must occur when pain or distress is beyond the level anticipated in the protocol description or when interventional control is not possible."* Veterinarians may make assessments of any reported health concerns and are available for consultation/training if needed.

RELATED GUIDELINES/POLICIES/SOP's
Veterinary Care Program
Health Monitoring, Record Keeping, and Communication for Rodents

Record Keeping for Research Animals: Experimental Procedures
Surgical Management, Monitoring and Record Keeping
OLAC SOP for Common Minor Medical Conditions and Treatments in Rodents

GENERAL CONSIDERATIONS

Daily health assessments will rely heavily on the Home Cage Assessment, as daily handling and restraint of rodents, especially mice, may cause unnecessary stress and/or behavioral changes that may impact breeding, experimental design, and overall well-being. If abnormalities are observed during the Home Cage Assessment, further assessment should be completed. While a complete hands-on assessment may not be required for all abnormalities, the guidelines below provide a framework for detailed health evaluations. The best time for OLAC staff to perform complete health assessments is during cage changing, while Lab Staff should perform complete health assessments any time they remove an animal from the cage for experimental handling. The recording of health assessments is only necessary when abnormalities are observed, or if health assessments are detailed in the approved animal protocol. Please see the "RELATED GUIDELINES/POLICIES/SOP's" listed above for further information on record keeping and communication.

1.0 Home Cage Assessment

Observing the animal(s) in the home cage will provide information about overall appearance and activity level, interaction with environmental enrichment, and behavior with respect to cage mates. Steps for Home Cage Assessment are as follows:

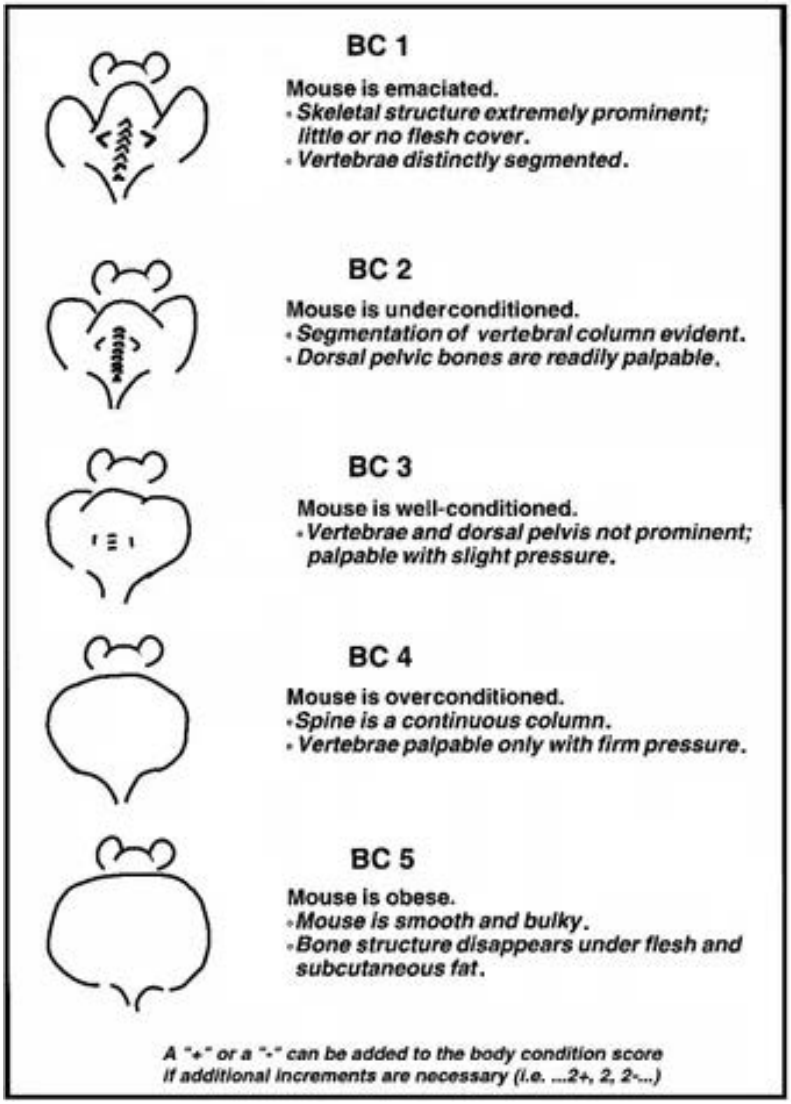
A. Read the cage card information so you know what is expected in the cage, i.e., strain, sex, age, number of animals.
B. If the animal(s) are known to have undergone a potentially painful procedure, try to observe the animal without moving the cage first, as rodents will mask pain, especially once disturbed by handlers. Signs of pain/distress can include lack of grooming, hunched or guarded posture, inactivity, etc. Standardized scoring methods such as Mice and Rat Grimace Scales and Body Condition Scores may be very useful in establishing assessments of pain/distress. Developing and assigning a study specific pain scale can be an effective tool to facilitate communication between observers and can serve as a convenient more objective method to determine when an animal's condition is deteriorating or has reached a clinical end point.
C. Next, move the rodent cage so it is at eye level. Start assessing the cage immediately as some health conditions, such as certain types of seizures, may only occur immediately after the cage is disturbed and will not be evident by the time the cage is moved to the change station or bench.
D. Rodents are generally active and inquisitive and can be seem moving about the cage and interacting with cage mates, perhaps eating, drinking or grooming. Behavioral indicators of a welfare issue can be obvious and include wounds, limping, hunched posture, dull or sluggish movements, large or

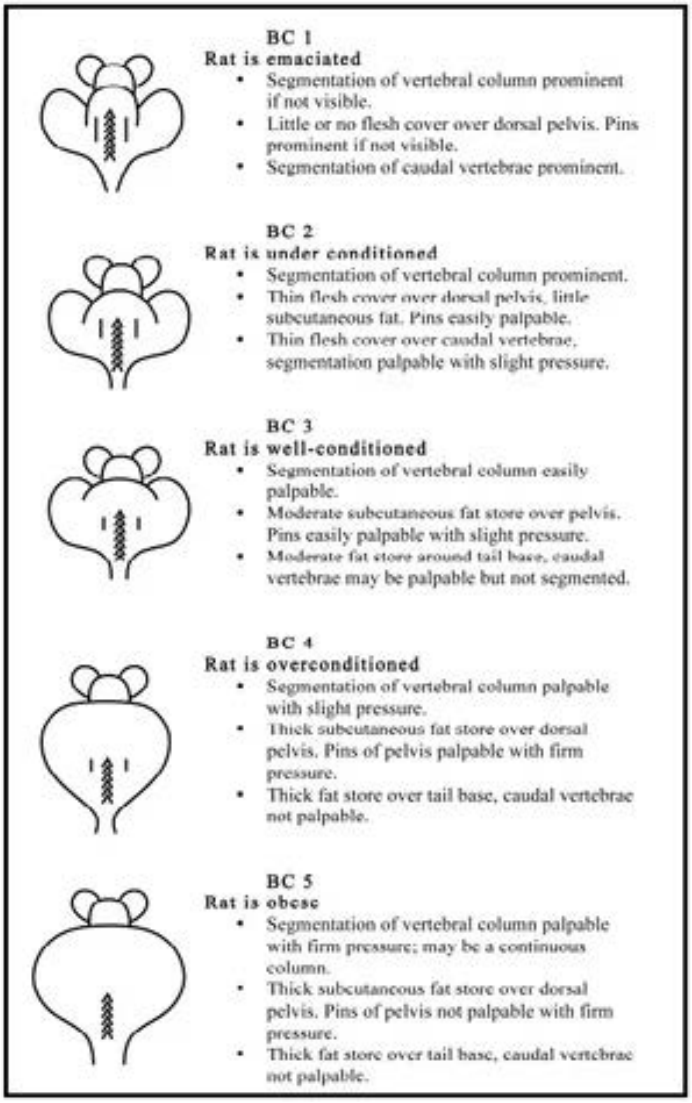
open tumors, or a mouse that does not move when the cage is manipulated (lethargy). Many behaviors, including indicators of pain, are more subtle and non-specific and will take practice and time to evaluate. Becoming familiar with normal appearances and behavior for the specific species, strain, etc., will aid in making valuable observations.

E. If any indicators of a welfare issue are observed upon home cage assessment, the cage should be pulled to make a more complete, hands-on assessment.

2.0 Hands-On Assessment

A. Using gentle species appropriate restraint, run a finger over the animal's coat to feel for any wounds or masses that may not have been evident while the animal was moving about the cage. Check hydration by tenting the skin between the shoulder blades. In a well hydrated animal, the skin should quickly return to its original shape. Use gentle palpation along the spine and hips to check body condition and score it from a 1 to 5 where 1 is emaciation and 5 is obese.





B. Examine each animal from nose to tail on both dorsally and ventrally.
    a. **Face and mouth.** Evaluate the eyes, ears, face and neck for abnormalities. Examine the incisors for malocclusion. Evaluate the color of the mucous membranes, which should be pink. Very pale or bluish mucous membranes are a clinical finding that should be reported ASAP.
    b. **Feet and limbs.** Examine the feet and limbs. Explore any gait abnormalities observed during the home cage exam. Note any sores, swelling in the hocks, or obvious lack of grip strength, etc.
    c. **Genital abnormalities**. Evaluate the female mammary chain for masses, abnormalities around the nipples or irregularities around the vulva. Examine the male penis by gently sliding back the prepuce. A purple or distended penis during exam may indicate a urinary

obstruction, which is a painful and life-threatening condition. Check the rectal area for swelling, trauma or prolapsed tissue.

d. **Abdominal palpation**. Palpate the abdomen by gently compressing the contents between the fingers from just under the ribs down to the hips. Common abnormalities palpated in the cranial aspect of the abdomen include tumors of the liver and spleen. In pregnant females, the distended uterus can be palpated in the mid abdomen and can extend up under the rib cage. In older females or retired breeders, masses in the mid abdomen may be due to uterine tumors. The bladder can be felt in the caudal abdomen. Mice will generally urinate as a stress response when picked up. A large or distended bladder can indicate an obstruction. In males, the glands of the reproductive organs can become enlarged and distended with fluid as the mice age which can be palpated in the caudal abdomen.

3.0 Common Health Conditions of Laboratory Mice and Rats

- Fight wounds: Most commonly seen in co-housed male mice, especially from strains such as BALB/c, SJL, and FVB. The typical presentation is a cluster of wounds on the rump, hips, and/or genital region, which may extend to the trunk of the body or forelegs.
- Neurologic conditions: Present in many ways including ataxia, head tilt, spinning when lifted by the tail, circling with inability to straighten out the path, and seizures. The primary clinical concern is ability to reach feed and water.
- Alopecia: Especially when seen in patches around the face or in one location on several mice within a group, is a sign of what is termed “barbering”. The skin is generally healthy, and short stubby hairs may be seen in the alopecic area. Unless there is secondary ulceration or inflammation of the skin, no medical treatment is necessary.
- Dermatitis: Includes ulcerative dermatitis. Erosions of the skin or small raised scabbed lesions may be seen. Describe the area and depth of the lesions and if there is any redness, discharge, scarring, etc.
- Tail lesions: May present as dermatitis, as fight wounds, or as concentric rings with hyperkeratosis. The latter has historically been related to very low humidity and is known as “ringtail”. Granulomas may form after tail biopsies for genotyping of mice.
- Mammary tumors: May be observed almost anywhere on the trunk of the body due to the extensive distribution of mammary tissue in females. These are subcutaneous, may be smooth or rough, and are usually easily moveable under the skin.
- Masses/Tumors: These should be evaluated with consideration of humane endpoints. Their size, location, secondary effects, etc. will be considered in decisions to observe, treat, or recommend euthanasia.

- Abscesses: Can occur in any location, but the most common are related to bite wounds, necrotic tumors, or blocked ducts to normal exocrine glands such as the preputial glands of male mice. They are usually a soft to firm swelling which may or may not be inflamed.
- Lymphadenopathy: Usually noted under the legs or in the neck region, but may occur anywhere lymph nodes are found, including within the abdomen. It may indicate a primary lymphoma or be an indication of systemic inflammatory responses.
- Microphthalmia or anophthalmia: Congenital conditions (common in C57BL/6 mice) which present as a partially opened or closed eye, or missing eye(s). Often tear production continues with poor drainage resulting in a mild watery or waxy ocular discharge. Most mice are stable and groom to keep the area clean, precluding the need for treatment.
- Conjunctivitis: Presents as swollen pink to red tissue under the eyelid and often a thick ocular discharge. It may be caused by foreign bodies such as a piece of bedding or be related to trauma to the conjunctiva or the globe.
- Keratitis: Inflammation of the cornea presents as a cloudy or vascular surface of the eye, and is often combined with conjunctivitis. Lack of tear production or inability to close the eyelids properly can lead to drying of the corneal surface. Ulceration of the cornea may be secondary to drying, or a result of trauma or displacement of the lens of the eye. A dent in the cornea may be visible and a tissue plug may be present if all layers of the cornea have been penetrated. The cornea is well innervated, so corneal lesions have potential to be quite painful.
- Cataracts: Seen as a central white material behind a clear cornea. Generally, they do not cause any problem for the animal, however occasionally the lens will luxate leading to inflammation within the eye, glaucoma, or even expulsion of the lens.
- Retro-orbital tumors, blood clots, or abscesses: Result in bulging of the eye forward of the normal position, and often difficulty in closing the eyelids.
- Arthritis: Presents as swelling and often redness of joints, with favoring of the affected limb or a reluctance to move. This may be transient, related to the strain or study, or a result of trauma.
- Respiratory Distress: including dyspnea, shallow rapid breathing, gasping, or abdominal effort in breathing. Nasal discharge may be seen dried on the nostrils, or a crusty material or porphyrin staining (in rats) may be seen around the eyes, nose and on the forelegs from grooming.
- Hydrocephalus: A congenital condition in which fluid builds up in the ventricles of the brain and does not distribute normally between the brain and spinal cord. Visibly these mice have a large, rounded head and shortened muzzle. They are smaller than littermates, and with time develop lethargy and neurologic abnormalities.
- Malocclusion: Rodent teeth grow throughout life. The teeth should meet in such a way that they grind on each other and on the feed to keep the teeth a normal length. When this does not happen teeth may grow into the palate or out of the

mouth making eating or drinking difficult for the animal. Deformities may be caused by congenitally defective jaw structure, damage to the developing teeth, or trauma to the mouth or jaw.

- Runt pups: Very small, poorly developing pups usually indicate a genetic abnormality, or are specific to a strain or experimental protocol.
- Imperforate vagina: A defect that may occur in the maturing young female mouse. It is produced by lack of opening of the vaginal membrane and appears as swelling between the anus and genital papilla giving the appearance of a male mouse.
- Dystocia: Difficulty in delivery of pups. Signs of dystocia include a pup visible in the vaginal canal but not passing, immobility and dehydration, distension of the abdomen with little muscle tone, or labor for an extended period of time (more than a couple hours).
- Prolapse of vaginal or uterine tissue: May be secondary to vaginal hyperplasia, or excessive abdominal contractions.
- Prolapsed penis (paraphimosis): Occurs in male mice when the penis is not retracted into the surrounding prepuce. The usual presentation is a swollen, dragging penis, often with secondary trauma to the surface skin. Blockage of the urethra may also be noted.
- Perineal cysts: The bulbo-urethral glands may become filled with fluid giving the appearance of a severely enlarged scrotum or perineum. Needle aspiration yields a clear slightly yellow fluid.
- Diarrhea: Noted as liquid feces when the animal is picked up or seen in the cage bedding.
- Ascites: The buildup of fluid in the abdomen may be induced by a study; in which case the inclusion of endpoint guidelines is critical. It can also indicate organ failure of the heart or liver, neoplasia, or lymphatic malfunction.
- Rectal prolapse: A bulging of the distal colon out of the rectum, common in mice affected by Helicobacter spp. or intestinal parasites; but also caused by straining, constipation, or unspecific reasons.

4.0 Experimental and Humane Endpoint Criteria

The experimental endpoint of a study occurs when the scientific aims and objectives have been reached. The humane endpoint is the point at which pain or distress in an experimental animal is prevented, terminated, or relieved. When there is potential for animal pain, distress, or suffering, investigators need to clearly outline the research objectives and procedures for assessing animal health. It is necessary to define appropriate experimental and humane endpoints that allow for early intervention (e.g., antibiotics, analgesics, or euthanasia), while attaining experimental objectives, minimizing data loss, reducing animal suffering, and improving the quality of data collected. Definition and use of effective animal health assessments is critical to effective observation and communication of experimental and humane endpoints. These assessments should be detailed in the approved animal protocol.

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