critical care management for laboratory rats

introduction

The laboratory rat continues to be broadly studied as a model species for investigating disease pathophysiology. Rats are second only to laboratory mice in the number used for biomedical research; fortunately, due to their similarities, many treatment applications described for mouse models can be extrapolated for administration to rats. Three main advantages to using laboratory rats for experimental purposes for studies are their comparatively larger size, coupled with their 3-week gestation period and production of large numbers of offspring. In addition, there are known scientific areas in which the laboratory rat is more similar to humans than the mouse, including the vascular system, the complexity of the rat brain for study of cerebral disorders, and the enzymatic ability of the rat liver to metabolize drugs. The continuing increase in rat genetics data and the rat genome have led to centralization of this information; these resources are highlighted further in Chapter 5. General information on working with laboratory rats is best reviewed in the companion text The Laboratory Rat (Sharp and Villano, 2012). Further background information on strains, stocks, and genotypes can also be obtained by visiting the originating vendor source websites, and additional resources are highlighted in Chapter 5.
When assessing laboratory rats, as described for laboratory mice, it will be essential to compile a thorough database of information on health status, research project enrollment, and any potential procedures or treatments already administered. Additional routine aspects of any critical care “history” (see details in Chapter 1) should include the background strain, gender, and age to gain the greatest portfolio of information prior to finalizing differential diagnoses. Further, any changes to the animal’s environmental and housing parameters should be reviewed for contribution to the clinical signs. These can include macroenvironmental influences of lighting, noise and vibration, and temperature and humidity of the room; as well, the microenvironment of the cage (diet, water source, housed singly or with other rats, bedding substrate) is to be considered with respect to maintenance of animal health.

If rats present in a critically poor state, as determined by the listing of abnormal health conditions in Chapter 1 (see Table 1.2), it will be essential to minimize stress and prioritize clinical interventions into smaller diagnostic and treatment steps. A medical record information template and sick animal reporting sheet are provided and can be used for any rats noted to be in less-than-optimal health (see Chapter 1, Figures 1.1 and 1.2). Typical values for biologic parameters in rats are presented in Table 3.1. The size of the average adult laboratory rat can range widely depending on gender, with females typically lighter weight than males (overall ranging from 250 to greater than 500 g). Despite the increased body size when compared to laboratory mice, there still are strong limitations on the ability to precisely quantify temperature, heart rate, and respiration rate without the use of telemetry or other special equipment in rats.

Knowledge of the appearance of a laboratory rat in clinically normal health will be key to ensure recognition of abnormal clinical signs should they appear. Visual examination of the animal is the first
step in assessing the overall physical condition of the laboratory rat and should be done with the rat in its home cage or housing setup prior to manual examination. Rats in critically poor health may benefit from access to supplemental heat and increased oxygen (flow rate 1–2 L/min) exposure (Klaphake, 2006).

As with standard handling practices for laboratory animals, to prevent transmission of potential human pathogens and unwanted exposure to animal allergens, fresh disposable gloves should be donned prior to manual restraint and handling of laboratory rats. Handling of rats throughout their time in the research environment will assist with their acclimation to this interaction with personnel. Data on a handling approach called “tickling” suggest that stress associated with handling and intraperitoneal (IP) injections is minimized using this technique (Cloutier et al., 2010). Playful handling includes gentle manipulation and petting of the rats for a few minutes, both before and after a procedure, for the patient to develop further acclimation to its human handlers.

Rats are difficult to lift by the scruff and will often vocalize and struggle against this type of restraint. Instead, a firm grip on the tail base of the rat will facilitate lifting the animal out of the cage (Figure 3.1), followed by placement of the animal on a stable surface

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**Table 3.1: Miscellaneous Parameters for the Laboratory Rat**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lifespan</td>
<td>2.5–4.0 years</td>
</tr>
<tr>
<td>Puberty</td>
<td>50 ± 10 days</td>
</tr>
<tr>
<td>Gestation</td>
<td>21–23 days</td>
</tr>
<tr>
<td>Male body weight</td>
<td>450–520 g(^a)</td>
</tr>
<tr>
<td>Female body weight</td>
<td>250–400 g(^a)</td>
</tr>
<tr>
<td>Blood volume</td>
<td>57–70 ml/kg = 17.1–21 ml total/300-g rat</td>
</tr>
<tr>
<td>Food intake</td>
<td>5–6 g/100 g BW/day</td>
</tr>
<tr>
<td>Water intake (ml/100 g BW/day)</td>
<td>10–12 ml/100 g BW/day</td>
</tr>
<tr>
<td>Packed cell volume (PCV)</td>
<td>35–57%</td>
</tr>
<tr>
<td>Glucose</td>
<td>80–300 (mg/dl)(^b)</td>
</tr>
<tr>
<td>Body temperature (rectal)</td>
<td>35.9–37.5°C (96.9–99.5°F)</td>
</tr>
<tr>
<td>Respiratory rate</td>
<td>70–150 breaths per minute</td>
</tr>
<tr>
<td>Heart rate</td>
<td>250–600+ beats per minute</td>
</tr>
</tbody>
</table>


\(^a\) Weights will vary depending on diet, age, stock, or strain.

\(^b\) Enzyme values are dependent on collection method and may be influenced by anesthesia.
Handling allows for the ability to closely observe skin and hair coat conditions, any ocular discharge or abnormalities, tooth overgrowth, abnormal masses, or unusual presentations in the anogenital region. Physiological aspects, like body weight (BW), activity, and behavior assessments, are useful to measure and monitor serially. Heart and lung sounds should be auscultated and can be performed using a pediatric stethoscope. Hair coat quality should be reviewed regarding location of areas of alopecia (baldness), open or closed wounds, or poor grooming. In addition, respiratory status (difficult or labored breathing with a more frequent/diminished rate than expected) should be evaluated. Relative perfusion status, ascertained by the color of mucous membranes (and potentially by color check of ear and tail tissue), reflects the transport of fluid and oxygen in blood to meet metabolic needs. Collectively, these physiologic measures

Fig. 3.1 Retrieval of rat from cage can be conducted using a firm grip at the tail base (A) to lift the animal and then placing it into the crook of the staff member’s arm (B). Alternatively, the rat is placed on a firm surface or benchtop and can be calmed by covering the eyes with a towel, which also serves to protect the handler from animal bites (C). The two-handed method of restraint allows for a partner to administer a physical examination or treat with therapeutics (D). (Images courtesy of University of Michigan, ULAM.)
provide a crude assessment of the “ABCs” (airway/breathing/circulation) of critical care medicine.

Rats in a critical state may need to be sedated to perform physical assessments while minimizing stress responses. Gentle palpation of the abdomen, using a pincer technique with the thumb and forefinger, should help to confirm pregnancy in females and further identify abnormalities like abdominal masses and growths, mammary tissue enlargement, or bladder distention. Finally, the particular experimental use of the rat, as described and approved in the approved proposal to the IACUC (Institutional Animal Care and Use Committee), must be considered, and any adverse effects of the experimental procedures should be documented.

**Body Condition Scoring**

Assessing general body condition, as described in mice, remains an excellent semiquantitative tool to apply toward rats for assessing health status. The use of a body condition score (BCS) scale (generally on a range from 1, for wasted or emaciated, to 5, for obese) is greatly enhanced by the incorporation of available cartoon diagrams that represent each score on the scale (Hickman and Swan, 2010). The uniformity of the diagrams can be exceptionally valuable for assessments done by a laboratory animal group with variable levels of experience in working with rats (Figure 3.2).

Overall percentages of weight loss should be tracked in rats yet may or may not indicate a loss of health condition, depending on the disease model and whether the animals are expected to develop spontaneous or experimental tumors or other syndromes. Typically, weight loss of more than 20–25% from preexperimental baseline may warrant critical care measures and potentially euthanasia, depending on institutional policies.

**Clinical Assessments of Ill Health and Pain in Rats**

Rats, though predators of some animals, are considered as prey species in the biomedical research environment. As such, similar to mice, they are conditioned to suppress overt painful behaviors, particularly when being handled. The following are clinical assessments of ill health and pain in rats (Kirsch et al., 2002, Kohn et al., 2007, Miller and Richardson, 2011, Roughan and Flecknell, 2004):

- Vocalization, particularly when handled or a painful area is palpated
BC 1
**Rat is emaciated**
- Segmentation of vertebral column prominent if not visible.
- Little or no flesh cover over dorsal pelvis. Pins prominent if not visible.
- Segmentation of caudal vertebrae prominent.

BC 2
**Rat is underconditioned**
- Segmentation of vertebral column prominent.
- Thin flesh cover over dorsal pelvis, little subcutaneous fat. Pins easily palpable.
- Thin flesh cover over caudal vertebrae, segmentation palpable with slight pressure.

BC 3
**Rat is well-conditioned**
- Segmentation of vertebral column easily palpable.
- Moderate subcutaneous fat store over pelvis. Pins easily palpable with slight pressure.
- Moderate fat store around tail base, caudal vertebrae may be palpable but not segmented.

BC 4
**Rat is overconditioned**
- Segmentation of vertebral column palpable with slight pressure.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.

BC 5
**Rat is obese**
- Segmentation of vertebral column palpable with firm pressure; may be a continuous column.
- Thick subcutaneous fat store over dorsal pelvis. Pins of pelvis not palpable with firm pressure.
- Thick fat store over tail base, caudal vertebrae not palpable.

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**Fig. 3.2** Schematic for scoring of the rat body condition. (Reprinted with permission from AALAS. Hickman, DL, and Swan, M. 2010. *J Am Assoc Lab Anim Sci* 49:155–159.)
• Reduced grooming or piloerection, leading to a “ruffled fur” appearance
• Reduced level of spontaneous and exploratory (sniffing, rearing) activity to the point that rats may not be moving (“moribund condition”)
• Hunched posture with “guarding” of abdomen and reduced mobility
• Squint-eyed appearance (either unilateral or bilateral)
• Increased aggressiveness on handling; may bite
• Porphyrin secretions (located around the eyes and nose); distinguish from bloody discharge by use of black light exposure to the secretion type (porphyrin will “glow”; blood will not)
• Distanced from cage mates
• Reduced body condition, likely secondary to reduced nutritional intake or experimental model resulting in muscle wasting and weight loss
• Self-mutilation (excessive licking, biting, scratching) of the painful area
• Abdominal writhing, increased back arching, falling or staggering, poor gait, and twitching
• Palpation of unexpected masses

**Monitoring Frequency**

Similar to the laboratory mouse, a detailed and descriptive plan for scheduled monitoring of rats both before and after any planned experimental procedures, including the provision of therapeutic treatments and supportive care, should be included in the IACUC protocol submission. Investigators should be aware that as the potential for pain/distress in research animals rises, there should be an increasing intensity of monitoring and frequency of observations.

**Objective Scoring Systems**

Professional and clinical judgments are essential for the evaluation of an animal’s well-being and are critical to the ultimate decision of euthanasia for humane reasons. As well, objective data-based approaches to predicting imminent death, when developed for specific experimental models, should facilitate the implementation of
timely euthanasia before the onset of clinically overt signs of moribund state (Toth and Gardiner, 2000). As described for mice, scoring systems are one way in which rats can be monitored throughout an experiment, and systems can be developed for individual experimental needs.

Novel approaches to pain assessment in laboratory rats have been described based on coding of facial expressions, referred to as the Rat Grimace Scale (RGS) (Langford et al., 2010, Sotocinal et al., 2011). Rats are the most common animal model for preclinical pain research (Mogil, 2009), and the RGS was used to improve quantification of pain in three common algesiometric assays: intraplantar instillation of complete Freund’s adjuvant, intra-articular kaolin/carrageenan administration, and laparotomy. In contrast to the grimace scale in mice, control rats display distinct bulging of the nose and cheek regions; with pain, the bridge of the nose flattens and elongates, further causing the whisker pads to flatten. This action unit of “nose/cheek flattening” shows the highest correlation with the presence of pain in the rat. The other action units, measured on the 0–2 scale, include orbital tightening and ear and whisker changes (Figure 3.3).

Overall, quantifying pain by facial changes provides a practical clinical assessment in that it can be performed in real time by trained investigators, animal technicians, and veterinary staff (Sotocinal et al., 2011).

**Veterinary Care Measures**

**Administration of Fluids**

Dehydration is often present in rats that have pain or are unwell and may be assessed by performing a skin tent or gentle pinch of scruff over the scapulae of the rat and assessing the time that passes for skin to return to normal placement. A prolonged return time indicates a degree of dehydration that should be ameliorated. Fluid administration through the subcutaneous (SC) route should be the least-invasive way to provide supplemental fluid support to the sick rat. Injections into the peritoneal cavity have been evaluated to determine how best to avoid accidental puncture of the cecum, and it has been shown that by avoiding the left lower side of the abdomen and injecting into the right lower side, the cecum is not affected (Coria-Avila et al., 2007). Intravenous (IV) injections

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can be performed for rats using the femoral, jugular, or tail vein, with animals appropriately sedated for access to the larger vessels (Figure 3.4); incisions may be required to gain access to the vessel of choice (Turner, Brabb, et al., 2011). Attempts at refinements for smaller-volume dosing have identified the superficial penile vein of the rat as an option for intravenous injections (Shapiro et al., 2010). As a reminder, the beneficial effect of playful handling (tickling) for rats is strongest when provided both immediately before and after injection (Cloutier et al., 2010).

Water and fluid replacement sources are gaining in popularity, expanding from products initially developed as sustainable fluid sources for the duration of rodent shipping and transport. The provision of these water replacements, in disposable single-use containers, is typically done on the cage floor for rapid access by those animals in ill health. These supplementary fluid sources, when combined

Fig. 3.3 Representative photographs of certain action units of the Rat Grimace Scale for a rat at baseline (facial grimacing not present, 0; a rat with moderate facial grimacing, 1; and a rat with obvious facial grimacing, 2) (Reprinted with permission from Biomed central open access. Sotocinal, SG, Sorge, RE, Zaloum, A, Tuttle, AH, Martin, LJ, Wieskopf, JS, Mapplebeck, JC, Wei, P, Zhan, S, Zhang, S, McDougall, JJ, King, OD, and Mogil, JS. 2011. Mol Pain 7:55.)
with food, can maintain the health of rodents for several days in the absence of routine water sources (Luo et al., 2003). Additional critical care considerations for nutritional support, fluid administration, and available products are provided in Chapter 4.

**Blood Sampling**

Blood sampling, or venipuncture, choices in rats may be influenced by sampling site, anesthetic agent, and method of collection (Fitzner Toft et al., 2006). Sampling allows for testing of serum chemistry parameters, as well as complete blood counts. Suggested sampling sites and further commentary are provided in Table 3.2.

As a guide, the volume of blood taken during a single survival collection should be limited to that needed, not in excess of 10% total blood volume (TBV) in rats; this also may be defined as a limit of about 1.0 ml/100 g BW (Sharp and LaRegina, 1998). For example, for 1% of BW to be withdrawn, 2.5 ml could be sampled at a single time point from a 250-g rat. Following sampling of 1% BW volume, replacement fluid therapy (0.5–1.0 ml SC or IP of sterile isotonic fluid) should be provided.

Retro-orbital blood sampling may be performed with animals under anesthesia but has been associated with subsequent lens opacities and a higher outcome of clotted samples, as compared to other methods (Mathieu, 2011). Other alternative sampling sites in rats include the lateral saphenous vein (Figure 3.5), the sublingual vein, and tail vessels.
The sublingual vessel can be accessed with the animal unanesthetized and securely restrained, similar to a basic hold used for performing an oral gavage. The mouth will open wide enough to expose the sublingual vasculature, and the oral cavity should be rinsed gently with saline or water and dried prior to sampling. Using a 25- or 23-gauge needle, the vein is punctured, and blood is collected via drip method into the appropriate collection tube. Gauze can then be packed under the tongue to achieve hemostasis (Kohlert, 2012). Care must be taken with any method to ensure that structures surrounding the sampling site are not injured. As well, digital pressure should be applied to achieve hemostasis following blood collection. For critically ill animals, the tail sectioning method may be used by making a transverse perpendicular incision at the tip of the tail (Liu et al., 1996).

The submandibular technique initially performed in mice has been adapted for use in rats. Rats should be lifted by the scruff tightly behind the ears to include as much loose skin as possible; alternatively, sedation will assist with the ability to limit mobility for this procedure. Once lifted by the scruff, the insertion point for the lancet should be located on the jawline, directly below the lateral canthus of the eye. Lancet sizes vary, but a 5.5-mm lancet has been used successfully in rats for this collection method. As with all sampling approaches, hemostasis must be achieved following sampling, typically through manual pressure over the lancet site of insertion (Arzadon, 2011).

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**Table 3.2: Recommended Sampling Sites and Related Information for Blood Collection in Rats**

<table>
<thead>
<tr>
<th>Anatomical Site</th>
<th>Anesthesia?</th>
<th>Approximate Range of Volume Collected</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lateral saphenous vein</td>
<td>Not required</td>
<td>Up to 1% of BW</td>
<td></td>
</tr>
<tr>
<td>Sublingual vein</td>
<td>Not required</td>
<td>50–100 µl</td>
<td></td>
</tr>
<tr>
<td>Lateral tail vein</td>
<td>Not required</td>
<td>Up to 1% of BW</td>
<td></td>
</tr>
<tr>
<td>Tail clip</td>
<td>Recommended</td>
<td>Up to 1% of BW</td>
<td>&lt;2 mm of distal end of tail should be clipped; analgesia should be considered</td>
</tr>
<tr>
<td>Jugular vein</td>
<td>Recommended</td>
<td>Up to 1% of BW</td>
<td></td>
</tr>
<tr>
<td>Submandibular</td>
<td>Not required</td>
<td>Up to 1% of BW</td>
<td></td>
</tr>
<tr>
<td>Retro-orbital vasculature</td>
<td>Required</td>
<td>Up to 1% of BW</td>
<td></td>
</tr>
<tr>
<td>Cardiac</td>
<td>Required</td>
<td>3+ ml</td>
<td>Terminal procedure only</td>
</tr>
</tbody>
</table>

*Source*: Modified from University of Pennsylvania, ULAR.
Jugular venipuncture may also be utilized, and it has been performed successfully in conscious and sedated animals using one-handed restraint of the rat. If sedating the animal, the thorax should be shaved to access the jugular vein and then swabbed with an alcohol pad. The collection needle should be inserted above the nipple line at a 20° angle using a minute vacuum, until blood is drawn into the collection tube. It is recommended to avoid continuous pressure on the syringe plunger so the vessel will not collapse. Gauze should

Fig. 3.5 Lateral saphenous vein sampling in the rat. The exterior leg is shaved delicately with a scalpel blade, and lubricant is applied over the vessel to allow blood to bead for collection (top); the vessel is pierced with a medical lancet and blood allowed to pool over the vessel (middle); the hematocrit tube is used to directly capture drops of blood for later submission for serum chemistry and complete blood counts (bottom).

Jugular venipuncture may also be utilized, and it has been performed successfully in conscious and sedated animals using one-handed restraint of the rat. If sedating the animal, the thorax should be shaved to access the jugular vein and then swabbed with an alcohol pad. The collection needle should be inserted above the nipple line at a 20° angle using a minute vacuum, until blood is drawn into the collection tube. It is recommended to avoid continuous pressure on the syringe plunger so the vessel will not collapse. Gauze should...
be held in place over the blood draw site until hemostasis is achieved (Zeleski et al., 2011).

**Body Temperature Monitoring**

Often, simple handling of ill rats will provide some indication of whether they are excessively cool or warm to the touch, but body temperature variations have to be extreme for manual detection. Rectal thermometer probes require gentle placement and positioning during procedures and may be more readily utilized in sedated rats, given their larger body size compared to mice. Microchip transponders that provide identification as well as thermometry are also useful for rodents (Bio Medic Data Systems, Seaford, DE).

Body temperature monitoring is critical for animals that are scheduled to undergo prolonged anesthesia; the goal is to mitigate hypothermia associated with experimental and surgical procedures. Suggestions to ameliorate hypothermia would include incubators and warm water bags, as well as Mylar-backed drapes, to reduce radiant heat loss. In addition, warm water recirculation or forced-air (Bair Hugger®, Arizant Healthcare, Eden Prairie, MN) blankets may be beneficial and synergistically effective when coupled with Mylar-backed draping (Koch et al., 2008).

No matter the type of draping used, personnel should ensure that draping allows for viewing of animals to ensure appropriate anesthetic administration and respiratory monitoring. Adverse incidents involving unrecognized surgical fires occurring below the level of a typical blue surgical drape have been described for the rat (Caro et al., 2011). Drapes coupled with a heat-emitting gel pad (Figure 3.6) can provide acceptable thermal support in the rat (Taylor, 2007).

**Bone Marrow Access**

Bone marrow aspiration from rats has been described as a method to obtain antemortem cell samples. A minimally invasive approach harvesting marrow from the femur (Figure 3.7) spares the knee joint and serves to minimize potential damage to the musculature of the quadriceps (Ordodi et al., 2006).

**Endotracheal Intubation**

Endotracheal intubation (Figure 3.8) can be readily performed in the rat using either a method of blind access or a strong external light
Fig. 3.6 Reusable heating pad (SnuggleSafe® Microwave Heatpad, West Sussex, UK) used for thermal support of anesthetized rats, with manufacturer’s cover intact. (Reprinted with permission from AALAS. Taylor, DK. 2007. J Am Assoc Lab Anim Sci 46:37–41.)

Fig. 3.7 Bone marrow harvesting in an anesthetized rat. In preparation for bone marrow harvesting, the rat should be anesthetized and intubated; then, the thigh area should be shaved and disinfected for the procedure (top). A 14-gauge needle and a 2-ml syringe are the required instruments for harvest; the needle pierces the anterior face of the thigh above the knee joint and is advanced into the femur prior to aspiration of cell sample (bottom). (Reprinted by permission from Macmillan Publishers Limited. Ordodi, VL, Mic, FA, Mic, AA, Tanasie, G, Ionac, M, Sandesc, D, and Paunescu, V. 2006. Lab Anim (NY) 35:41–44.)

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source that penetrates the skin to illuminate the larynx and facilitate intubation. Other options for endotracheal tubing can be fashioned from standard 2-ml syringes and a light source to illuminate the oropharyngeal cavity, providing easy localization of the larynx (Molthen, 2006, Ordodi et al., 2005). If the rat is in respiratory distress, intubation should be undertaken with caution; however, it can be attempted in animals weighing more than 100 g (Paul-Murphy, 1996). Note that in the critically ill rat, intubation may be challenging and should be

**Fig. 3.8** Intubation in the rat. (A) This photograph of a laryngoscope with a light source incident on the proximal end illustrates how light is transmitted to the distal surfaces. (B) Image of laryngeal opening of a rat showing the epiglottis (red arrowhead), arytenoid cartilages (black arrows), and caudal margin of the soft palate (black arrowhead). Visual appearance is similar in the mouse. (C) An anesthetized rat positioned and restrained on inclined plane. (D) The laryngoscope is positioned in the oral cavity to provide visualization of the larynx. The tongue is grasped against the shaft of the laryngoscope. The stylet and tracheal tube are shown before being inserted into the oral cavity. Note the relative position of stylet within the tracheal tube. (Reprinted with permission from AALAS. Molthen, RC. 2006. *J Am Assoc Lab Anim Sci* 45:88–93; and Rivera, B, Miller, S, Brown, E, and Price, R. 2005. *Contemp Top Lab Anim Sci* 44:52–55.)
attempted only as a last resort to gain airway access if tracheostomy cannot be performed (see relevant section in Chapter 4).

**Injections and Oral Administration**

Injections can be performed routinely using multiple routes (Table 3.3) for the rat, including subcutaneous (SC), intradermal (ID), intraperitoneal (IP), intratracheal (IT), and intravenous (IV), as described in Chapter 1. Intravascular access ports, typically placed surgically in the subcutaneous space over the shoulder area for laboratory rats, can be accessed using Huber needles for substance administration (Figure 3.9). The intramuscular route (IM) can also be more readily used with small-volume injections, as compared to the mouse.

**Table 3.3: Recommendations for Injection Dose Limits Based on Weight of Laboratory Rats**

<table>
<thead>
<tr>
<th>Route</th>
<th>PO</th>
<th>SC</th>
<th>IP</th>
<th>IM</th>
<th>IV (Bolus)</th>
<th>IV (Slow)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dose (ml/kg) Weight (kg)</td>
<td>Injection Limits (ml/kg)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dose (ml/kg) Weight (kg)</td>
<td>10</td>
<td>5</td>
<td>10</td>
<td>0.1</td>
<td>5</td>
<td>20</td>
</tr>
<tr>
<td>0.200</td>
<td>2.0 mL</td>
<td>1.0 mL</td>
<td>2.0 mL</td>
<td>0.02 mL</td>
<td>1.0 mL</td>
<td>4.0 mL</td>
</tr>
<tr>
<td>0.225</td>
<td>2.2</td>
<td>1.1</td>
<td>2.2</td>
<td>0.07</td>
<td>1.12</td>
<td>4.5</td>
</tr>
<tr>
<td>0.250</td>
<td>2.5</td>
<td>1.2</td>
<td>2.5</td>
<td>0.07</td>
<td>1.25</td>
<td>5.0</td>
</tr>
<tr>
<td>0.275</td>
<td>2.7</td>
<td>1.3</td>
<td>2.7</td>
<td>0.07</td>
<td>1.35</td>
<td>6.0</td>
</tr>
<tr>
<td>0.300</td>
<td>3.0</td>
<td>1.5</td>
<td>3.0</td>
<td>0.07</td>
<td>1.5</td>
<td>6.0</td>
</tr>
<tr>
<td>0.325</td>
<td>3.2</td>
<td>1.6</td>
<td>2.5</td>
<td>0.07</td>
<td>1.6</td>
<td>6.5</td>
</tr>
<tr>
<td>0.350</td>
<td>3.5</td>
<td>1.7</td>
<td>2.5</td>
<td>0.07</td>
<td>1.75</td>
<td>7.0</td>
</tr>
<tr>
<td>0.375</td>
<td>3.7</td>
<td>1.8</td>
<td>2.5</td>
<td>0.07</td>
<td>1.85</td>
<td>7.5</td>
</tr>
<tr>
<td>0.400</td>
<td>4.0</td>
<td>2.0</td>
<td>2.5</td>
<td>0.07</td>
<td>2.0</td>
<td>8.0</td>
</tr>
<tr>
<td>0.425</td>
<td>4.2</td>
<td>2.1</td>
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*Source: Modified from University of Pennsylvania, ULAR.*

*a* A bolus is a larger dose given over a shorter period of time.

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Administration of drugs and fluids can also be done by oral gavage; however, animals must be healthy enough to drink from syringes. This method of fluid/drug administration is less invasive and is easily undertaken once rats are trained to the syringe (Figure 3.10) (Atcha et al., 2010, Turner, Brabb, et al., 2011). If repeated oral (PO) dosing is required, acclimation of rats to handling and to gavaging (with up to 5 ml/kg of a control aqueous material) will help to diminish chronic stress from the procedures (Turner et al., 2012).

Creative approaches to disguising medications in palatable substances, like analgesics in gelatin desserts (Jell-O®), have been described for rats (Flecknell, Orr, et al., 1999, Flecknell, Roughan, et al., 1999). Attention should be paid to the differences in dosing that may be required if delivering oral medications compared to subcutaneous administrations; as well, rats may need to acclimate to the novel substance prior to drug incorporation (Martin et al., 2001).

**Urine Sampling**

Clinically healthy rats will often dribble urine, which allows for a free-catch sample (Klaphake, 2006). Slight pressure can be applied...
over the bladder to assist with expression of urine, and one should ensure that an appropriate sterile receptacle is positioned to collect the sample (Kurien et al., 2004). Critically ill rats should be stabilized prior to attempting urinary catheterization if urine collection by other methods has been unsuccessful. Urinary catheterization should only be performed on anesthetized animals. Aseptic technique (see Chapter 4, “Perioperative Care Considerations”) and an atraumatic approach should be used during placement of a urinary catheter. Prior to insertion of the catheter, the external urinary orifice should be gently cleansed using a disinfecting (e.g., chlorhexidine) solution. The individual performing the catheterization is advised to don sterile surgical gloves, use a sterile catheter, and apply a small amount of sterile water-soluble lubricant on the external urinary orifice. Additional sterile lubricant should be applied in a thin layer to cover the surface of the urinary catheter for ease of insertion into the urinary orifice, as described for mice (St. Claire et al., 1999). The diameter of the urinary catheter should be the minimum that can be inserted into the bladder and still prevent urinary leakage around the catheter.

The anatomy of the female rat is unique in that the urinary orifice is external and just anterior to the vaginal opening. Adult female rats can be catheterized with number 50 polyethylene (PE) tubing

Fig. 3.10 Oral dosing of laboratory rats. Rats readily drinking galantamine (0.5 mg/kg) by the novel syringe-dosing method after an acclimation training period (left); animal voluntarily consuming nutritional supplement from a syringe (right) (Reprinted with permission from AALAS. Atcha, Z., Rourke, C., Neo, A.H., Goh, C.W., Lim, J.S., Aw, C.C., Browne, E.R., and Pemberton, D.J. 2010. *J Am Assoc Lab Anim Sci* 49:335–343; and Turner, P.V., Brabb, T., Pekow, C., and Vasbinder, M.A. 2011, *J Am Assoc Lab Anim Sci* 50:600–613.)
(2.9 French), a 3.5-French TomCat catheter, or a number 4 Coude urethral catheter that has a bend to the tip of the catheter. This bend facilitates passage of the catheter through the urethra. A guidewire can be threaded through the PE tubing to increase the rigidity of the catheter. Care should be taken that the tip of the guidewire does not extend past the end of the catheter. Guidewires can be made of stainless steel surgical wire and coated with a water-soluble lubricant to ease placement and removal from the PE tubing. The approximate distance from the external urinary orifice to the neck of the bladder for 200-g female rats is 17 mm (St. Claire et al., 1999).

If urine concentration tests are to be performed in rats, personnel should be aware that dehydration may be secondary to any prolonged water deprivation necessary for this type of assay. Studies that assessed clinical condition and BW, at a frequency of every 2 h beginning 16 h after food and water deprivation were initiated, showed a mean BW loss of 8% at 16 h and nearly 10% at 22 h. Clinical dehydration was noted by 22 h, whereas appropriate urine concentration was noted at 16 h. Therefore, it is recommended to complete the rat urine concentration test within a 16-h period to maintain welfare of the animals for this procedure (Kulick et al., 2005).

abnormal, critical, and emergent conditions

Categories of laboratory rodent health concerns are discussed in alphabetical order to facilitate location by the reader. Under each topic, “cause and impact” has been provided, and “potential treatments” offer suggestions about procedures, therapeutic treatments, or husbandry and environmental alterations. Every attempt has been made to provide citations from the literature for evidence-based medical outcomes.

It is essential to note that certain abnormal conditions can be assessed and treated in laboratory rats by similar methods to those done for laboratory mice; see relevant sections in Chapter 2 for the following:

- Abdominal swelling
- Abscessation
- Cage flooding
- Cannibalization
- Cross fostering

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• Dystocia
• Fight wounds
• Fractures/orthopedic problems
• Hemorrhage
• Mortality (sudden death)
• Ocular lesions
• Respiratory distress
• Trauma
• Ulcerative dermatitis

For those health concerns that list drug therapy options, please refer to the rodent formulary provided in Appendix C for additional details on dosages and route of delivery.

Burns

• Cause and impact: Burns may be the unfortunate outcome of improper surgical preparation of the animal with alcohol-based disinfectants. Care should be taken to ensure that animal skin that is prepped with alcohol is not then inadvertently ignited by cautery tools during surgery. Smoke inhalation and superficial and partial-thickness burns have been documented to result from this sort of accident (Figure 3.11), the severity of which may be missed due to obstructive surgical draping (Caro et al., 2011). Burns may result in blistering and skin necrosis, shock, and secondary bacterial infections.

• Potential treatments: Animals should be provided with oxygen supplementation and stabilized following smoke inhalation. The extent of burn damage should be assessed and any wounds cleansed, debrided, and covered with a topical antibacterial cream, like silver sulfadiazine. To prevent secondary bacterial infection, treatment with systemic antibiotics should be considered. As well, warmed subcutaneous fluids should be provided to offset shock and prevent dehydration. Pain management should be a priority, with opioids or nonsteroidal anti-inflammatory drugs (NSAIDs) provided throughout the duration of the initial healing phases. The prognosis will depend on the extent and thickness of the burned area; aggressive management and monitoring are advised, and euthanasia may be warranted.
Catheter Infections

- **Cause and impact:** Rats, more commonly than mice, are catheterized using vascular access ports (Figure 3.9), typically into the jugular vein, for chronic administration of any variety of test substances. Any indwelling catheter has the potential to serve as a nidus of infection, leading to systemic illness with clinical signs of decreased body condition and activity and altered behavior. As well, localized inflammation can occur at the skin surface and in the subcutaneous space with development of pustular material and a threat to catheter patency, particularly during chronic studies.

- **Potential treatments:** Frequent attention to catheter care is the key prevention strategy against infections. Catheters should be flushed at least twice weekly, once at the time of treatment and again 3 days later. Skin overlying the vascular access port can be cleansed with a chlorhexidine scrub, alternated with dilute povidone–iodine solution. Gentle manual restraint of the rats will permit access to the port site; a noncoring Huber needle can then be inserted through the skin and into the port reservoir. It is recommended to flush the catheter with a volume of about 0.2 ml saline, followed by 0.2 ml heparinized dextrose. Following treatment administration, an additional 0.2 ml saline can
be injected to purge the catheter line. Catheters should be “locked” with an anticoagulant to assist with patency; this material can include heparinized dextrose or may involve 500 IU of heparinized glycerol (Wachtman et al., 2006, Weiner et al., 2012).

Anecdotally, flushing the catheter with saline every 3 to 7 days has been evaluated and found to have negative impacts on the ability to withdraw blood samples (Luo et al., 2004).

Should rats present with signs of systemic disease, antibiotics can be administered, along with supplemental fluids and potential removal of the catheter from the animal. If the rat is intended only for a study requiring chronic catheterization, it may be best to euthanize the septic rat in lieu of aggressive attempts at treatments.

**Malocclusion (Incisors) and Caries**

- **Cause and impact:** Incisor overgrowth may occur secondary to congenital tooth patterns or may relate to lack of appropriate caging materials for gnawing, softened foods, or a genetic predisposition. Rats that have difficulty prehending food will be anorectic, lose weight, and typically show a decreased BCS within a relatively rapid (~24- to 48-h) time frame.

  Dental caries may develop spontaneously in certain transgenic rat strains; animals should be monitored for signs of anorexia and potential pain secondary to development of caries (Nishijima et al., 2007).

- **Potential treatments:** Treatment includes trimming overgrown teeth to a normal length and alignment. A diamond blade, Dremel®, and dental burr are recommended tools for use on rat dentition. Care should be taken not to crack or split the teeth, which could potentially cause pain and lead to tooth root infections. For valuable rats with potential oral pain secondary to teeth abnormalities and caries, anesthetic extractions of affected teeth may be a potential treatment. Subsequent provision of softened nutritional supplements and wetted chow may be necessary to maintain body condition (see Chapter 4, “Nutritional Therapy Considerations”).

  Special attention should be given to the potential for malocclusion in aged rats (especially noted in Wistar rats) during
long-term rodent studies as the increased incidence may be detrimental to maintenance of health and general well-being (Dontas et al., 2010).

**Moribund/Weak/Paralyzed**

- **Cause and impact:** Hind limb weakness (paresis) and paralysis in laboratory rats are associated with trauma, weakness, and dysfunction of the musculoskeletal and nervous systems, neurologic disease models, adverse surgical outcomes, or trauma that may be due to environmental or experimental influence. Neoplasia and nonneoplastic diseases, such as osteoarthritis, bone fractures, or peripheral neuropathies, may also occur, particularly in aged rats (Ceccarelli and Rozengurt, 2002).

  Rat models of spinal cord injury are commonly implemented for biomedical research; in addition to the induced spinal cord lesions, the injured rats may experience alterations of the liver, lung, bladder, and kidneys (Robinson et al., 2012).

  Be aware that rats may also self-injure (autophagia or autotomy) as a consequence of spinal cord or peripheral nerve injury research, associated with altered mobility and pain (Figure 3.12).

- **Potential treatments:** Rats found in a weakened and potentially unresponsive state should be provided with ancillary and supportive care of warmed subcutaneous fluids (2–4 ml 0.9% NaCl) and softened bedding substrates, nutritional supplementation (including softened food pellets on the cage bottom), and supplemental heat, until the level of responsiveness is determined. It is critical to increase the frequency of monitoring and determine humane endpoints that eliminate prolonged suffering for paralyzed or moribund rats (see Chapter 4, “Humane or ‘Clinical’ Endpoint Considerations”).

  For those rat models of spinal cord injuries, researchers should be aware that suprapubic bladder catheterizations performed postinjury will not prevent development of renal abnormalities in rats; therefore, manual expression of the bladder should be performed two to three times daily to eliminate urine accumulations (Robinson et al., 2012).

  Increased observations and monitoring should be done for those animals with self-inflicted lesions. If autophagia has
resulted in limb injury, then degree of lameness, amount of swelling, and integrity of the wound should be assessed (Geertsema and Lindsell, 2011). Treatments should include analgesia, cleansing of the wound site, bandaging of the area following application of local analgesics and triple antibiotic ointments, and placement of a restraint collar to prevent the rat from accessing the injured area (see Chapter 4, “Restraint Collar Considerations”). Metronidazole can be applied (“painted”) topically over the area where self-injury has occurred due to its aversive taste; a NewSkin® bandage can then be painted over the metronidazole to prolong the presence of the drug on the skin and promote healing (Zhang et al., 2001). If the self-injury is severe to the point of severe welfare alterations, euthanasia is recommended.

More often than not, moribund and paralyzed animals will require euthanasia if there is no improvement or change in activity status within 24 h of initial presentation.

Fig. 3.12 Self-injury in an adult male Sprague-Dawley rat. Following a left sciatic nerve transection and 48 h of postoperative analgesia, this rat began to self-mutilate the left hind foot and toes (highlighted in the enlarged image on the right) within 3 days, despite preventive application of metronidazole/New Skin®. The animal was inhibited from doing further damage by the application of a bitter-tasting (Grannick’s Bitter Apple®) spray. (Images courtesy of University of Pennsylvania, ULAR.)
Ocular Lesions Secondary to Anesthesia

- **Cause and impact:** Cloudiness of corneal tissue can be secondary to application of anesthesia and omission of topical eye lubrication while animals are under anesthesia. Corneal lesions and keratoconjunctivitis sicca (Kufoy et al., 1989) can be more severe in animals anesthetized with ketamine plus xylazine; minimal ocular changes have been noted in rats following enflurane or isoflurane anesthesia. Corneal lesions can be observed within 24 h after injectable anesthetic administration and may be irreversible. Compared with Sprague-Dawley and Lewis rats, Wistar, Long-Evans, and Fischer 344 rats had increased incidence and severity of corneal lesions after anesthesia with ketamine plus xylazine, suggesting that these three strains are at increased risk for developing postanesthetic corneal lesions with this regimen (Turner and Albassam, 2005).

Acute reversible corneal lesions, attributable to a side effect of xylazine, have been documented in rats (Calderone et al., 1986).

- **Potential treatments:** Treatment with a sterile ophthalmic lubricant (e.g., PuralubeTM) can assist with the prevention of dry eyes and will soothe irritation. Ophthalmic products of this nature should be applied any time a rodent is under anesthesia to provide a protective film over the ocular surface, similar to what is done in routine veterinary clinical practice. The severity of corneal changes has been diminished in rats for which ketamine plus xylazine anesthesia was reversed with yohimbine (Turner and Albassam, 2005).

Topical ophthalmic ointment, with or without added antibiotics, is warranted as a first-line approach even for animals that appear to be otherwise behaviorally normal, despite the ocular lesion. Certain lesions will be painful, with notation of animals scratching at the eye and face; these animals should receive topical (proparacaine) drops and systemic analgesics (meloxicam 5 mg/kg SC) for pain relief. Surgical enucleation may be warranted for correction of severe ocular injuries in rats.

Poor Body Condition

- **Cause and impact:** Rats may present clinically with a thin, alopecic, and hunched appearance without much forewarning (Figure 3.13). This may be attributed to malocclusion (see
relevant section on this topic) that is preventing ingestion of hard food items (e.g., pelleted chow) or as a result of a gastrointestinal abnormality, husbandry alterations, experimental manipulation, or internal tumor burden (Mexas et al., 2011).

Differentials should include behavioral stereotypies that are preventing the animal from grooming or causing the animal to overgroom. Infectious agents should be ruled out as a cause of alopecia through the performance of skin scrapes and fungal cultures. If experimental treatments are potentially toxic or unpalatable, this should be further discussed with the research team.

- **Potential treatments:** The logical and prioritized causes should be treated first, typically including administration of a subcutaneous bolus of fluids, nutritional support, and heat supplementation if the animal is hypothermic. If the animals were expected to succumb to experimental disease, then aggressive treatment efforts should be aimed at assisting with nutritional supplements and comforting the animal with provision of cage enrichments and bedding substrates to attempt to reach the experimental endpoint.

The rest of the affiliated colony should be evaluated further to determine if the condition is endemic, and blood sampling can be done for further diagnostic assessments of potential infectious pathogens. Body condition scoring should be monitored daily and BWs checked routinely to track any further decline. Animals that become quiet, less alert, and
unresponsive should be considered for euthanasia prior to a moribund state and spontaneous death.

**Ringtail**

- **Cause and impact:** Ringtail is a pathologic condition of the tail, and sometimes feet, characterized by dry skin and annular constrictions that can result in necrosis and loss of portions of the tail (Figure 3.14).

  The cause of ringtail is not completely understood, although the condition is typically noted in weanling animals and may be caused by relative environmental humidity levels below 25%. Other contributing factors, such as dietary deficiencies, genetic susceptibility, environmental temperatures, and degree of hydration, also have been proposed. The variety of possible etiologic factors suggests that this syndrome might be the clinical expression of more than one causative agent or that more than one causative agent may be necessary to induce ringtail (Crippa et al., 2000).

- **Potential treatments:** Treatment with over-the-counter lanolin ointment (a nontoxic, inexpensive, and effective moisturizer) has been successful when initiated prior to the condition becoming severe enough that there is tail necrosis. It can also be applied prophylactically to rats starting at 7 days of age for groups that may have a history of disease (Taylor et al., 2006).

**Ulcerative Dermatitis**

- **Causes and impact:** Ulcerative dermatitis (UD) has been noted in Zucker lean rats, especially distal to forelimbs, with isolated lesions on the head and behind the ears. Determining the appropriate sensitivity profile to cultured bacteria is essential to provide an effective antibiotic treatment. Skin lesions may be secondary to dietary deficiencies, such as linoleic acid deficiency, with manifestation of focal areas of alopecia to diffuse areas of moist dermatitis on the head, face, ear pinnae, and neckline.

- **Potential treatments:** Administration of leptin topically at 5 µg daily to affected areas can provide reduction in wound size and severity. As well, trimming of hind toenails to prevent self-inflicted skin trauma is advisable (Oppelt, 2005).
Fig. 3.14 Examples of ringtail in preweanling rats. (A) Normal, healthy rat pup tail. This tail was given a condition score of 0. Note that the distal portion of the tail has been biopsied for genotyping. Representative clinical cases of ringtail of varying severity: (B) Rat pup tail showing some flaking of the skin with mild constrictions. This tail was given a condition score of 1. (C) Rat pup tail clearly exhibiting annular constrictions and some malformation of tail tissue. This tail was given a condition score of 2. (D) Rat pup tail exhibiting annular constrictions with some malformation of tissue; the tail tip appears necrotic. This tail was given a condition score of 3. Topical application of lanolin to tails appearing like those in Figure 3.14B and 3.14C returned them to a healthy and clinical normal tail appearance. Due to the level of necrosis in Figure 3.14D, one might consider amputation of the tail tip to remove necrotic tissue. (Reprinted with permission from AALAS. Taylor, DK, Rogers, MM, and Hankenson, FC. 2006. J Am Assoc Lab Anim Sci 45:83–87.)
Concurrent correction of dietary imbalances, topical application of betadine cleanses, triple antibiotic ointment, and zinc oxide may be beneficial (Godfrey et al., 2005).

For a comprehensive listing of various treatments of UD in laboratory mice that may be efficacious for UD in laboratory rats, refer to the relevant section in Chapter 2.

**Urolithiasis**

- **Cause and impact:** Clinical signs indicative of urolithiasis include combinations of hematuria, red-stained bedding with abnormal urine, red-stained or wet pelage (especially over the abdomen), sensitivity to touch in the abdominal area, swollen or palpable kidney or bladder, unkempt fur, anorexia, reduced urination, reduced water intake, and unexpected weight loss or gain (due to fluid retention) (Newland et al., 2005).

  Partial-to-complete obstruction of urinary outflow can cause mild-to-severe pressure necrosis of the renal pelvis, medulla, and eventually the cortex. In addition, urinary calculi can inflame and cause degeneration and necrosis of the epithelial lining of the urinary tract. Incomplete emptying of the urinary system due to obstruction, coupled with the loss of epithelial integrity, allows bacterial overgrowth and subsequently an ascending urinary tract infection. In the case of a severe infection, bacteria can gain access to systemic circulation and cause sepsis.

  Urolithiasis has also been linked to a model of lymphocytic choriomeningitis virus (LCMV) infection in Lewis rats (Mook et al., 2004).

- **Potential treatments:** These factors indicated above, when taken as a whole, make it clear that once potentially obstructive uroliths form, the future health of the rat is at considerable risk, perhaps irreversibly, because calculi are highly persistent.

  Diet may need to be altered if the rats are to be maintained in the research colony. For example, of those rats maintained on a purified American Institute of Nutrition (AIN)-93 diet, males are considerably more at risk for urolithiasis and develop the condition within a few months of eating the diet (Newland et al., 2005). As rats on the AIN-93 diet aged, the discrepancy in risk between males and females increased; in fact, by 100 weeks, nearly 60% of male rats died of urolithiasis.
three times the prevalence seen in female rats. Postmortem analyses suggested that males were more likely to have bladder calculi than were females, who usually formed calculi in the kidney. Euthanasia of rats with severe clinical signs of urinary dysfunction, likely secondary to stone formation, is warranted to limit continued discomfort and potential spontaneous mortality (Figure 3.15).

**research-related medical issues**

Additional topics concerning laboratory rat health felt to warrant further information herein due to their prevalence in contemporary research environments are provided next in alphabetical order. Under each topic, “background information” is provided, and “potential treatments” offer suggestions about procedures, therapeutic treatments, and further considerations.

**Arthritis Models**

- **Background:** Induction of arthritis to better investigate the pathogenesis of inflammation and test the potential
for antiarthritic agents is a classic model in the laboratory rodent. Differing models include adjuvant arthritis (typically in male Lewis rats, with injection at the tail base or into the foot pad); type II collagen arthritis (typically in female rats given bovine type II collagen); antigen arthritis; and injection of substances like capsaicin and carrageenan (Bendele, 2001). Tail and paw swelling with edema is expected as an acute inflammatory reaction, and the severity of paw swelling may render the animal immobile. While experimental treatments can be administered, these rats would typically be scientifically justified not to receive analgesics for pain management due to concern for impact on the development of experimental inflammation.

- **Potential treatments:** Provision of soft bedding and feeding of a softened food or nutritional supplement on the cage floor are recommended (Flecknell, 2001). Positioning of the water source, such that the animal can access fluids easily without additional pressure on the inflamed joints, should be considered. If analgesic treatments can be administered and not compromise the data, one could provide NSAIDs (indomethacin), dexamethasone or other corticosteroids (at low doses to avoid toxicity seen with chronic use), methotrexate (low dose), and biological agents like soluble tumor necrosis factor R2 (TNF-R2) that are currently marked for human treatments of arthritis (Bendele, 2001). It is not recommended that animals be handled daily as this may increase their stress and alter the desired inflammatory outcomes; instead, every-other-day (EOD) handling should be sufficient (Brand, 2005). Humane endpoints must be established for this type of model to best limit the duration and intensity of pain sensation (National Research Council [NRC], 2009).

**Cranial Implant Maintenance**

- **Background:** Neuroscience research often requires surgical implantation of an apparatus that permits direct manipulation of brain tissue or measurement of neuronal activity in conscious animals. Successful factors for longevity of cranial implants in rats have been described (Gardiner and Toth, 1999). Contributing factors include accurate targeting of the location of interest, aseptic surgical technique, maximal adherence of acrylic cement to the bone through proper
preparation of the skull surface, and provision of ventilation during the thermogenic phase of cement curing. For the skin to heal properly around the implant site, apposition of skin to the implant is essential to promote comfort and reduce the likelihood of secondary bacterial infections.

• **Potential treatments:** Wound margins should be treated topically and liberally with antibiotic ointment (daily for 7 days postsurgery) (Gardiner and Toth, 1999). Repositioning of the skin to adhere better to the headpiece may be of use for a nonhealing incision site, and skin retraction may need to be employed to gain the coverage needed over the skullcap. Systemic antibiotics can also be administered pre- and postsurgically.

Continued maintenance of the areas around the implant will likely be necessary to avoid the buildup of crusts and potential for secondary bacterial infections (Figure 3.16). Using a nonirritating antiseptic/cleansing solution, gently remove any scabbing and minimize disruption of wound margins. Trimming of hairs along the margin is useful to minimize irritation. Antibiotic ointment may then be applied as needed. Cultures should be routinely taken to track potential bacterial infections and provide relevant systemic antibiotics, if necessary.

![Fig. 3.16 Cranial implant complications in an adult rat. Porphyrin staining was noted around the eyes and nares, indicative of poor health and stress. Although the wound margins appeared relatively healthy, apposition was compromised. Due to declining health, this animal was euthanized, and *Pseudomonas aeruginosa* was cultured from the brainstem surface (arrow). (Images courtesy of University of Pennsylvania, ULAR.)](image)

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Incontinence Secondary to Spinal Cord Injury Models

- **Background:** Urine scald secondary to urinary incontinence from spinal cord injury studies can pose significant clinical problems. Urine scald is likely to cause discomfort, signified by skin with severe redness and warmth and the presence of urine or urine stains. Related complications can include intractable skin ulceration, secondary bacterial dermatitis, and self-trauma.

- **Potential treatments:** Amelioration of the discomfort has been attempted by application of a commercially available hexamethyldisiloxane (HMDS)-based skin protectant barrier film, 3M No-Sting Barrier Film® (3M Corporation, St. Paul, MN), which is used to treat diaper rash in human infants and urine scald in incontinent adults (St. Claire et al., 1997).

For incontinent rats, urinary bladders can be manually expressed every 8 h until rats are observed to urinate without assistance. After each timed expression of the urinary bladder, barrier film is applied to clean, dry skin by spraying a uniform coat of film over the affected area. Animals should be observed for signs of discomfort after application of the barrier film. In addition, daily monitoring for paralysis, signs of dehydration, food intake, and evidence and degree of urine scald should be performed. Skin irritation can be rated from minor (slight redness, cool to touch) to major (severe redness, warm to touch) with or without moisture from urine (St. Claire et al., 1997).

Treatment of spinal cord injury with minocycline, an antibiotic with neuroprotective effects, has been beneficial for restoration of motor coordination and hind limb reflexes. This antibiotic can be administered within 1 h after injury (90 mg/kg IP), followed by doses (45 mg/kg IP) twice daily for 5 days (Teng et al., 2004).

Middle Cerebral Artery Occlusion in Rat Models of Stroke

- **Background:** The major complication of the stroke model is the substantial morbidity and mortality that occurs postoperatively due to respiratory distress caused by stimulation of the sympathetic nerve system. Prolonged occlusion of the common carotid arteries can lead to prolonged tachycardia and potential for arrhythmias.
• **Potential treatments:** Administration of bupivacaine (0.25% solution 0.1–0.2 ml SC) can ameliorate left-side heart failure that would otherwise lead to mortality and may improve the outcome of the model (Wang Fischer et al., 2003). Scoring sheets have been developed for evaluation of pain management in this model as the administration of NSAIDs for pain relief may not be permitted due to bias of data outcomes (Kirsch et al., 2002).

Obese and Diabetic Rat Models

• **Background:** Rat models for obesity and diabetes research are beneficial to the identification of surgical and therapeutic interventions that can be translated to the related human disease syndromes. Performing surgery in physiologically compromised rats, particularly with their body conformation of a thinner thoracic cavity and larger abdominal mass, can result in adverse effects like hypoglycemia, difficulty with dehiscence of incision sites, and anesthesia reactions that can potentially lead to fatal outcomes.

• **Potential treatments:** For those rats undergoing surgery, attention to the minimal time for fasting, both presurgically and postsurgically, is key (see Chapter 4, “Fasting Considerations”). To prevent dehiscence of surgical sites, it is recommended to avoid surgical skin clips for this model and instead close incisions with continuous suture patterns, with minimal suture size, and subcuticular closure patterns. Maintaining the anesthetic dose of isoflurane to no more than 1.5%, with an oxygen flow rate of 0.5 L/min, facilitates recovery issues and maintains animals at a reasonable depth of anesthesia. As well, supplemental heat should be provided as described and endorsed for all surgical models in rodents. Application of these refinements has been shown to contribute to a survival rate of approximately 90% for gastrointestinal procedures performed in obese and diabetic rats (Baran et al., 2011).

Providing supportive care and specialized environments will be best for these obese models. For animals with evidence of diabetes, more frequent cage changes and provision of more absorbent bedding substrates should be used in the rat cages to compensate for increased urine production.
Opportunistic Infections in Immunodeficient Rat Models

• **Background:** Similar to cases in laboratory mice that are immunodeficient (see relevant section in Chapter 2), *Klebsiella oxytoca* has been identified as a monoculture from urogenital tract infections and abscesses, as well as serving as the etiology for otitis, keratoconjunctivitis, meningitis, lymphadenitis, and pneumonia (Bleich et al., 2008). Abnormal colonization with *K. pneumonia* has also been documented following antibiotic treatment in nude rats (Hansen, 1995). Rats enrolled in longevity studies may succumb to opportunistic infections with age, prior to collection of desired data points.

• **Potential treatments:** Husbandry and environmental changes have been useful in eradicating opportunists, like *Pneumocystis carinii*. Housing rats in autoclaved cages, with autoclaved bedding, and provision of trimethorim-sulfa-methoxazole-treated acidified water have minimized reported health issues. As well, providing a diet with 14% protein and 3.5% fat, along with pair housing of rats, has effectively extended the lifespan and improved overall health in aged rats (Zahorsky-Reeves et al., 2007). Also, nutritional supplementation in the form of sterile solidified gels may be provided (see Chapter 4, “Nutritional Therapy Considerations”).

Pododermatitis

• **Background:** Pododermatitis can be common in mature rats (>300 g) chronically housed (>1 year) in wire-bottom cages but is less commonly noted when animals are housed on bedding (Carraway and Witt, 2003, Peace et al., 2001). The problem is characterized by chronic, suppurative inflammatory lesions (ulcers) on the plantar surfaces of the hind feet; lesions may be reddened and raised, with keratinized growth developing into crusts and scabs (Peace et al., 2001, Sharp and Villano, 2012).

• **Potential treatments:** Topical and systemic treatment options may be limited by impacts on study data; however, antibiotics and analgesics would be ideal for addressing the infectious nature and associated pain from these lesions. Placement of some sort of flattened and softer bedding substrate or surface on the wire cage bottom, akin to sterile gauze squares (4 x 4 inches), has a significant preventive benefit for diminishing the potential for ulceration of noted foot sores.
(Dimeo and Mitchell, 2005, Peace et al., 2001). Soaking of affected feet (hydrotherapy) in Epsom salt solution (4 cups of warm water to 1 teaspoon of salt) has anecdotally been successful for resolution of surface infection and softening of crusts covering foot wounds. Surgical debridement is rarely successful, and prognosis for complete resolution is guarded (Langlois, 2004). Closely monitor affected animals to ensure that any rats experiencing severe pain and distress are removed from the study and euthanized.

**Spontaneously Hypertensive Rat Models**

- **Background:** To promote maintenance of BW of senescent female spontaneously hypertensive rats (SHRs), supplementing powdered feed is useful to offset loss of appetite and weight loss.

- **Potential treatments:** With age, SHR rats will benefit from the addition of powdered food to ensure that BWs remain stable and to prevent malnutrition that could lead to premature death. Rats were also given powdered rat chow in shallow bowls to facilitate the eating and digestion of food. As the female SHR matures, special care and handling are essential to help maintain BW and good health. With only modest changes in routine (i.e., powdered food) and an attentive eye on the rats’ daily activities, it is possible to maintain these rats in a healthy condition until the termination of the study (Belanger et al., 1999).

**Tumor Burden in Rat Models**

- **Background:** Rodent tumor models are quite common in laboratory animal facilities. Institutional guidance should be followed with respect to size of allowable tumors and increased monitoring of animal health (see Chapter 4, “Tumor Development and Monitoring Considerations”). Spontaneously occurring tumors may also develop and should be managed based on how the animal’s overall health and body condition fares.

  Tumors in rats may be secondary to foreign body reactions, particularly for intra-abdominal telemetry devices in certain
strains (Popovic et al., 2004). Tumor incidence should be considered an adverse outcome in instrumented rats.

Subcutaneous masses involving the mammary chain are usually benign fibroadenomas, with less than 10% being malignant. Mammary tissue in rats is extensive, and masses can occur anywhere from the neck to the inguinal region, arising in locations as dorsal as the flank areas and across the shoulders.

Paraneoplastic syndrome in young rats has been described secondary to extensive mammary neoplasia (Figure 3.17) (Mexas et al., 2011).

![Fig. 3.17](image)

**Fig. 3.17** Induced neoplasia model young (2-month-old) female rat. (Top) The animal was very thin with a BCS on initial examination at 1.5 of 5. (Bottom) Visible and firm palpable masses extended bilaterally throughout regions of mammary tissue on the ventral aspect of the mouse (highlighted in boxes). The animal had a 48-h history of lethargy and dehydration; on physical examination, the rat became extremely stressed, developed agonal breathing, and was euthanized immediately. Necropsy identified widespread mammary tumors (corresponding with palpated masses) and multiple organ abnormalities, including calcification as a paraneoplastic syndrome. (Images courtesy of University of Pennsylvania, ULAR.)
Fig. 3.18 Rat with a spontaneous ulcerated mammary tumor. Due to the ulceration and location in the left axillary region (left, ventral view; right, right-side recumbent view), this rat would require heightened monitoring for alterations to mobility, hemorrhage of the mass, and further irritation to the tumor site. (Images courtesy of University of Pennsylvania, ULAR.)

- **Potential treatments:** Surgical excision is the most common form of therapy and results in more cures than all other modalities combined (Mehler et al., 2004). Mammary gland tumor removal can be straightforward; in brief, the vascular supply to these tumors is limited and therefore can be ligated using vascular clamps or suture material. Once the neoplastic tissue is removed, the tissue space and subcutaneous tissue can be closed with 3–0 vicryl suture (using a simple-interrupted or continuous pattern). Overlying skin can be closed with suture, wound clips, or tissue glue (Fisher, 2002).

It may be possible to have the tumor treated in some other manner to continue using the animal in a study; however, overall animal welfare should not be compromised if the tumor is left untreated, affects mobility, or ulcerates (Figure 3.18). Tumor development may affect animal welfare for those animals in long-term studies and decrease confidence in the reliability of data outcomes from the model.

**euthanasia**

Euthanasia is the process of inducing painless death in animals. To the greatest extent possible, animals being euthanized should not experience pain, fear, or other significant stress prior to their death.
Carbon dioxide (CO₂) exposure or narcosis is a frequently used euthanasia method for small laboratory animals due to its rapid onset of action, safety, low cost, and ready availability. Exposure times for carbon dioxide differ dramatically depending on the age of the rat to be euthanized; rats older than 21 days typically require 5 min of exposure time (Pritchett-Corning, 2009). Injectable and inhalant methods are therefore preferred unless individuals have received hands-on training for physical methods of euthanasia. Further discussion is provided in Chapter 4, “Euthanasia Considerations” and in the AVMA Guidelines for the Euthanasia of Animals (American Veterinary Medical Association [AVMA], 2013).

references


Langford, DJ, Bailey, AL, Chanda, ML, Clarke, SE, Drummond, TE, Echols, S, Glick, S, Ingrao, J, Klassen-Ross, T, Lacroix-Fralish,


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Paul-Murphy, J. 1996. Little critters: emergency medicine for small rodents, pp. 714–718. Fifth International Veterinary Emergency and Critical Care Symposium, San Antonio, TX.


