

WAYNE STATE UNIVERSITY DIVISION OF RESEARCH	Division of Laboratory Animal Resources
	Guidance Document
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Veterinary Recommendations for Anesthesia and Analgesia

This Guidance Document is to establish recommendations for anesthesia and analgesia for use in research animals.

PRINCIPLES OF ANESTHESIA AND ANALGESIA

1. **The proper anesthetic and analgesic agents must be used in order to eliminate or reduce the potential for pain and distress during the peri-operative period.**
2. Withholding anesthesia or analgesia must be justified and approved in the IACUC protocol.
3. A multimodal approach to analgesia should be employed to offer the best broad-spectrum pain control possible. This includes the use of different categories of analgesics in combination to address different sources of pain perception/stimulation.

For example, a surgical procedure may use a local anesthetic block of lidocaine and bupivacaine at the incision site, and systemic administration of an NSAID for inflammatory pain and an opioid.

4. According to the 8th edition of the Guide for the Care and Use of Laboratory Animals (NRC), "Guidelines for the selection and proper use of analgesic and anesthetic drugs should be developed and periodically reviewed and updated as standards and techniques are refined."

ANESTHESICS

1. Inhalant Anesthetics
 - a. The inhalant anesthetics include gases such as isoflurane and sevoflurane. These anesthetics require an anesthetic machine set-up. In addition, use of a scavenger system is required to prevent personnel exposure to the waste anesthetics. For short procedures it may be possible to administer inhalant anesthesia via a drop jar. For this procedure a cotton ball or gauze soaked with the anesthetic is placed in a jar with the animal. This procedure must be performed under a fume hood, the animal CANNOT contact anesthetic, and can only be done for minor quick procedures. Animals will recover quickly after removal from jar.
 - b. Advantages: safe and reliable, predictable and rapid control of anesthetic depth, not controlled substances
 - c. Disadvantages: induction must be closely monitored, personal training, special equipment required, potential risk to staff (if not appropriately scavenged)
2. Injectable anesthetics
 - a. Injectable general anesthetics include ketamine/xylazine and pentobarbital. Most of the commonly used agents are administered via intraperitoneal (IP) injection (figure 4).
 - b. Local anesthetics are often delivered subcutaneously along the incision site. They could also be used in nerve blocks or epidural administration. Local anesthetics are not adequate as the only analgesic for any surgical procedure unless scientifically justified in the protocol.

- c. Advantages: They can be used without expensive supporting equipment such as the anesthesia machines required with the use of inhalants, they are easily transported, and are relatively inexpensive.
 - d. Disadvantages: Prolonged recovery times. The animal will have to metabolize the drug in order to completely recover from anesthesia. In addition, once the agent is injected, the anesthetic depth cannot be adjusted throughout the procedure except to achieve a deeper anesthetic plane by giving additional drug if the animal demonstrates signs of arousal. Animals which are sick or compromised may have a difficult time with these anesthetics due to changes in their ability to metabolize the drugs. Also, many of the commonly used injectable anesthetics are controlled substances which will require the laboratory comply with all rules regarding controlled substances and obtain a DEA license.
3. Monitoring Anesthetic Depth

Anesthetic depth should be gauged prior to conducting any surgical manipulation and throughout the surgical procedure. Loss of reflexes (e.g., pedal, corneal, palpebral) can be used to assess appropriate anesthetic plane. There should not response to toe pinch. Signs of Inadequate anesthetic depth include purposeful movement, reflexes present, response to painful stimulus, or twitching whiskers. Equipment (e.g. pulse oximeter) may also be used to monitor depth of anesthesia. Changes in heart rate, respiratory rate, or blood pressure may indicate whether an animal is at too light or too deep of an anesthetic plane. Depth of anesthesia should be assessed every 10-15 minutes during surgery.

ANALGESIA

1. Pain assessment and Analgesia

Assessing pain is difficult in animals. Some animals are species of prey and are adapted to hide signs of pain and distress. Clinical signs associated with pain are species specific, but some common signs of all species include changes in appearance such as hunched, scruffy, porphyrin staining (rats/mice), or changes in activity, including less active or inactive, hyperactive or pacing, abnormal postures such as back arching, belly pressing, wound guarding, or writhing. Also, decreased appetite, isolation from cage mates, exaggerated or decreased response to handling, vocalization can be used to gauge pain and distress. Recently, there has been some focus on assessing rodent facial expression or grimace scale in order to assess pain (Figure 1). It is important to pay close attention to the animal's appearance and behavior post-surgery in order to observe subtle changes that may indicate the need for additional pain management.

Analgesia should be administered to **ALL** post-surgical animals unless otherwise justified in the protocol. If there is concern regarding an animal's clinical condition post-surgery and additional analgesia is necessary, contact DLAR veterinary staff for further guidance on treating the animal.

Whenever possible multimodal analgesia is recommended. This involves providing a more 'balanced analgesia' through multiple methods or modalities. Local anesthetics at the incision site are often used in conjunction with a stronger opioid or NSAID analgesic.

RECOMMENDATIONS BY PROCEDURE

See specific doses, routes, and frequencies in the formularies below.

1. Rodent laparotomy (example major surgery)

Anesthesia induction via isoflurane in an induction chamber at 3-5% followed by isoflurane via facemask at 2-4%. Buprenorphine or other moderate to strong analgesic is given immediately after induction prior to patient prep to allow adequate time to reach therapeutic levels. Subsequently, moderate to strong analgesia is administered at appropriate intervals for 48 hours minimum post-operatively. Rodents are monitored closely thereafter to evaluate for signs of pain, and additional analgesia is given until evidence of pain is no longer present.

2. Rodent subcutaneous implant or vascular catheter placement (example minor surgery)

Anesthesia induction via ketamine-xylazine. Carprofen is given immediately after induction prior to patient prep to allow adequate time to reach therapeutic levels. Carprofen is given as directed for 24 hours minimum post-operatively. Rodents are monitored closely thereafter to evaluate for signs of pain, and additional analgesia is given until evidence of pain is no longer present.

3. Swine laparotomy

Anesthesia induction via ketamine-acepromazine. Pre-emptive analgesia is given immediately after induction prior to patient prep to allow adequate time to reach therapeutic levels; buprenorphine (for moderate pain; time to effect - 30 minutes IV & 60 minutes IM) and NSAID (time to effect - 60 minutes IV or IM). Following endotracheal intubation, anesthesia is maintained with isoflurane. Local anesthetics, lidocaine and bupivacaine, are given SC at the location of planned incision. Post-operative analgesia, buprenorphine and NSAID, is given at appropriate intervals for at least 48 hours postoperatively. Swine are monitored closely postoperatively to evaluate for signs of pain, and additional analgesia is given until evidence of pain is no longer present.

4. Dog thoracotomy

Anesthesia induction via ketamine-diazepam. Pre-emptive analgesia is given immediately after induction prior to patient prep to allow adequate time to reach therapeutic levels; buprenorphine (for severe pain; time to effect - 30 minutes IV & 60 minutes IM) and NSAID (time to effect - 60 minutes IV or IM). A fentanyl transdermal patch is secured on skin (time to effect - 12-24 hours). Following endotracheal intubation, anesthesia is maintained with isoflurane. Local anesthetics, lidocaine and bupivacaine, are given SC at the location of planned incision. Once skin incision is made, an intercostal nerve block is performed. Post-operative analgesia is given at appropriate intervals for at least 72 hours postoperatively, buprenorphine for 12-24 hours until fentanyl takes effect, fentanyl transdermal patch in place for 72 hours, and NSAID for 3-5 days. Dogs are monitored closely postoperatively to evaluate for signs of pain, and additional analgesia is given until evidence of pain is no longer present.

Neonatal Rodents (mice and rats)

Neonatal rodents must have adequate anesthesia and analgesia when undergoing surgical procedures. It is important to balance safety with effectiveness when using anesthetics in neonatal animals. Neonates have an immature hepatic/renal system which can lead to prolonged anesthesia and a narrow margin of safety when using injectable medications. Inhalant anesthetics (isoflurane/sevoflurane) or hypothermia (<7 days of age) are the recommended methods of anesthesia in neonatal rodents. Opioid analgesics have been used successfully in neonatal rodents. However, these drugs should be administered at the lower end of the published dose range to avoid complications.

Hypothermia Anesthesia	
Age	Altricial rodents up to 7 days old
Induction	2-4 minutes. Protect pup by placing into finger of a glove or paper lined test tube. This will avoid skin damage caused by direct contact with ice/cold water. Immerse pup in ice water or place on crushed ice (2-3°C or 35-37°F). Observe pups closely during induction.
Maintenance	Up to 15 minutes anesthesia. Remove pups from ice bath when adequate anesthesia is achieved (immobile/lethargic). The pup can be maintained by placing on an ice pack covered with latex/paper towel.
Recovery	Up to 1 hour. Avoid rapid warming during the recovery period. Recommend re-warming using an incubator set at 90-95°F (32-25°C) or in a paper-lined cage set over a circulating warm water blanket. Electric heating blankets and heat lamps are not recommended.
Additional considerations	Fiber optic lighting should be used during surgical procedure to help maintain hypothermia (incandescent lamps can warm surgical field). Steps should be taken to avoid rejection by dam post-surgery. Recommend: remove blood/disinfectants from pup after surgery (wipe with wet gauze and dry), make sure neonate is completely recovered (warm, pink, breathing, moving), place neonates in bedding/nesting material from home cage to obtain appropriate scent, and return neonates as a group to home cage.

FORMULARY FOR MOST COMMONLY USED SPECIES AND DRUGS

Mouse and Rat Formulary						
Analgesics	Dose (mg/kg)		Route	Freq	Comments	
	Mouse	Rat				
Acetaminophen Oral dose		100-300	PO	q4h	Not adequate as a sole analgesic except for very minor pain. May be combined with another class of analgesic for post-op pain.	
Water bottle	1-2 mg/ml drinking water	6 mg/ml drinking water	PO		Change water every other day. Water must be placed on cage 48 hours prior to painful procedure in order for rats to acclimate.	
Recommended: Buprenorphine	0.05 – 0.1	0.01 – 0.05	SC	q12h	Excellent for moderate to severe pain.	
Carprofen	5	5	SC	q24h	Good for mild to moderate pain.	
Meloxicam	1-5	1-2	PO, SC	q24h		
Tramadol	20 - 40	5 - 20	IP		For chronic or severe pain. Literature does not define recommended dosing interval in rodents. In other species administered up to TID. Start at BID dosing.	
Anesthetics	Mouse	Rat	Route	Duration	Comments	
Recommended: Isoflurane	2-5%	2-5%	Inhalation		Gold standard anesthetic. Must have appropriate equipment to use safely (precision vaporizer and scavenging).	
Ketamine / xylazine	90-120 10	40-80 5-10	IP	30-45 min	Ketamine combinations are the next best anesthetic if isoflurane cannot be used. Do not re-dose xylazine ; if additional dose needed provide ~1/3 dose ketamine	
Ketamine / medetomidine	50-75 1-10	60 0.4	IP			
Ketamine / xylazine / acepromazine	100 2.5 2.5	40 8 4	IM, IP			
Pentobarbital	30-90	30-60	IP	60-120 min	Not readily available.	
Tribromoethanol	250	Not recomm- ended	IP		Very short term anesthesia, not recommended for survival procedures; inappropriate storage or mixing may result in toxicity.	
Reversal Agents	Mouse	Rat	Route	Duration	Comments	
Yohimbine	0.2	0.2	IP	NA	Reverses xylazine	
Atipamazole	1	1	SC	NA	Reverses xylazine and medetomidine	
Local anesthetics	Mouse	Rat	Route	Duration	Comments	
Bupivacaine / Lidocaine mixture	1.5 mg/kg	1.5 mg/kg	SC	4-8 hrs	Slow onset, long duration	Mix together in same syringe for infiltration around incision
	0.5 mg/kg	0.5 mg/kg		<1 hr	Rapid onset, short duration	

Dog Formulary				
Analgesics	Dose (mg/kg)	Route	Frequency	Comments
OPIOIDS				
Recommended: Buprenorphine	0.01-0.03	IV/IM/SC	Q6h	Mild to moderate pain
Fentanyl transdermal	<10kg 25 µg/h 10-25kg 50 µg/h >25kg 100 µg/h	dermal patch	q72h	Severe pain; 24 hours for maximal effect
NSAIDS/OTHER				
Recommended: Carprofen	2.2 4.4	PO/IV/SC	Q12h Q24h	Excellent synergistic effect with opioids
Meloxicam	0.2 mg/kg once, then 0.1 mg/kg	IV/SC/PO	q24h	Excellent synergistic effect with opioids
Acetaminophen	10-15	PO	q6-8h	Antipyretic, mild pain
Anesthetics	Dose (mg/kg)	Route	Frequency	Comments
INHALATION				
Recommended: Isoflurane	3-5% induction 1-3% maintenance	Inhalation	continuous	Must use precision isoflurane vaporizer
Sevoflurane	5-7% induction 2-4% maintenance	Inhalation	continuous	Must use precision sevoflurane vaporizer
INJECTABLE				
Ketamine / diazepam	5.0 mg/kg 0.25 mg/kg	IV in same syringe	As needed	Use for anesthetic induction; duration 20-30 minutes
Ketamine / midazolam	5.0 mg/kg 0.25 mg/kg	IV in same syringe	As needed	Use for anesthetic induction; duration 20-30 minutes
Propofol	5.5-8.0 mg/kg* = induction 0.4 mg/kg/min* = maintenance	IV IV	Slowly to effect Continuous rate infusion (CRI)	Must administer slowly over 30-60 seconds to avoid apnea, hypotension. Rapid recovery in 10-20 minutes
Pentobarbital*	25-30 mg/kg	IV	Slowly to effect; single or intermittent bolus	Single dose duration approximately 45 minutes
Local anesthetics	Dose (mg/kg)	Route	Frequency	Comments
Bupivacaine / Lidocaine mixture	1.5 mg/kg 0.5 mg/kg	SC	Once	Mix in same syringe for infiltration or local nerve block. Bupivacaine has slow onset (20-30 minutes), longer duration (4-8 hours); Lidocaine has rapid onset (5-10 minutes), shorter duration (1-2 hours).

Dog Formulary				
Neuromuscular Blocking	Dose (mg/kg)	Route	Frequency	Comments
Pancuronium	0.05-0.1 mg/kg, then 0.01 mg/kg	IV	Once, then as needed	Duration of effect 30-45 minutes
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Atropine Neostigmine methylsulfate	0.02 mg/kg 0.04 mg/kg	IV	Once No more than 3 times	Reverses neuromuscular block; give atropine first

Swine Formulary ³				
Analgesics	Dose (mg/kg)	Route	Frequency	Comments
OPIOIDS				
Recommended:				
Buprenorphine	0.01-0.05	IV/IM/SC	q8-12h	Mild to moderate pain
Butorphanol	0.1-0.3	IM/SC	q4-6h	Mild to moderate pain
Fentanyl transdermal	5 µg/kg/h	dermal patch	q72h	Severe pain; dosage variable depending upon breed, age, patch location, heat, moisture, procedure
NSAIDS				
Recommended:				
Carprofen	2.0 3.0-4.0	PO/IV/SC/I M	q12-24h q24h	Excellent synergistic effect with opioids
Meloxicam	0.1 mg/kg 0.4 mg/kg	PO IV/SC	q24h	Excellent synergistic effect with opioids
Anesthetics	Dose (mg/kg)	Route	Frequency	Comments
INHALATION				
Recommended:				
Isoflurane	3-5% induction 1.5-3% maintenance	Inhalation	continuous	Must use precision isoflurane vaporizer
Sevoflurane	5-7% induction 3-4% maintenance	Inhalation	continuous	Must use precision sevoflurane vaporizer
INJECTABLE				
Ketamine/ acepromazine	22-33 mg/kg 1.1 mg/kg	IM/SC	once	Use for anesthetic induction; duration 30 minutes; slightly cardio-depressant
Ketamine/ midazolam	33.0 mg/kg 0.5 mg/kg	IM/SC	once	Use for anesthetic induction; duration 45-60 minutes; profoundly hypothermic due to peripheral vasodilation; terminal procedures only
Ketamine/ xylazine	20 mg/kg 2 mg/kg	IM/SC	Once	Use for anesthetic induction; use anticholinergic to reverse cardiodepression/heart block
Telazol® (tiletamine/zolazepam)	2.0-8.8 mg/kg	IM/SC	Once	20 minutes immobilization; use for minor surgery & anesthetic induction; hypothermia, cardiodepression
Telazol®/ xylazine	4.4 mg/kg 2.2 mg/kg	IM/SC	Once	Use for anesthetic induction and intubation; not recommended for cardiovascular studies

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Swine Formulary ³				
Propofol	0.83-1.66 mg/kg induction 14-20 mg/kg/min* maintenance	IV IV	Slowly to effect Continuous rate infusion (CRI)	Must administer slowly over 30-60 seconds to avoid apnea, hypotension. Effective sedation and muscle relaxation with minimal cardiodepression at lower dosages; poor analgesia at lower dosages
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Yohimbine	0.05-0.1 mg/kg	IV slowly	Once	Reverses sedative, cardiodepression, analgesic effects of xylazine
Local anesthetics	Dose (mg/kg)	Route	Frequency	Comments
Bupivacaine / Lidocaine mixture	1.5 mg/kg 0.5 mg/kg	SC	Once	Mix in same syringe for infiltration or local nerve block. Bupivacaine has slow onset (20-30 minutes), longer duration (4-8 hours); Lidocaine has rapid onset (5-10 minutes), shorter duration (1-2 hours).
Neuromuscular Blocking	Dose (mg/kg)	Route	Frequency	Comments
Pancuronium	0.02-0.15 mg/kg	IV	Once, then as needed	Duration of effect 30-45 minutes
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Atropine Neostigmine methylsulfate	0.02 mg/kg 0.04 mg/kg	IV	Once No more than 3 times	Reverses neuromuscular block; give atropine first

Cat Formulary				
Analgesics	Dose (mg/kg)	Route	Frequency	Comments
OPIOIDS				
Recommended: Buprenorphine	0.01-0.03	IV/IM/SC PO (sublingual)	q6h q6-12h	Mild to moderate pain
Fentanyl transdermal	25 µg/h	dermal patch	q72h	Severe pain; 6-12 hours for maximal effect
NSAIDS				
Recommended: Meloxicam	0.1 mg/kg IV/SC/PO once, then 0.05 mg/kg PO	IV/SC/PO	q24h	Excellent synergistic effect with opioids; chronic dosing PO only
Anesthetics	Dose (mg/kg)	Route	Frequency	Comments
INHALATION				
Recommended: Isoflurane	3-5% induction 1-3% maintenance	Inhalation	continuous	Must use precision isoflurane vaporizer

Cat Formulary				
Sevoflurane	5-8% induction 2-4% maintenance	Inhalation	continuous	Must use precision sevoflurane vaporizer
INJECTABLE				
Ketamine/ diazepam	5.0 mg/kg 0.25 mg/kg	IV in same syringe	As needed	Use for anesthetic induction; duration 20-30 minutes
Ketamine/ midazolam	5-10 mg/kg 0.2-0.4 mg/kg	IM in same syringe	As needed	Use for anesthetic induction; duration 20-30 minutes
Ketamine/ xylazine	10 1	IM	Once	Administer xylazine first as stimulation of chemoreceptor trigger zone frequently causes vomiting; wait 10 minutes, then administer ketamine
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Yohimbine	0.05-0.1 mg/kg	IV slowly	Once	Reverses sedative, cardiodepression, analgesic effects of xylazine
Local anesthetics	Dose (mg/kg)	Route	Frequency	Comments
Bupivacaine / Lidocaine mixture	1.5 mg/kg 0.5 mg/kg	SC	Once	Mix in same syringe for infiltration or local nerve block. Bupivacaine has slow onset (20-30 minutes), longer duration (4-8 hours); Lidocaine has rapid onset (5-10 minutes), shorter duration (1-2 hours).
Neuromuscular Blocking	Dose (mg/kg)	Route	Frequency	Comments
Pancuronium	0.05-0.1 mg/kg, then 0.01 mg/kg	IV	Once, then as needed	Duration of effect 30-45 minutes
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Atropine Neostigmine methylsulfate	0.02 mg/kg 0.04 mg/kg	IV	Once No more than 3 times	Reverses neuromuscular block; give atropine first
Edrophonium	0.5 mg/kg	IV	Once No more than 5 times	Reverses neuromuscular block; complete reversal takes 5-45 minutes

Rabbit Formulary				
Analgesics	Dose (mg/kg)	Route	Frequency	Comments
OPIOIDS				
Recommended: Buprenorphine	0.01-0.05	IV/SC	q6-12h	Mild to moderate pain
Fentanyl transdermal	½ 25 µg/h patch for 3 kg rabbit ¹	dermal patch	q72h	Severe pain; do not cut patch, cover ½ not in use
NSAIDS				
Recommended: Meloxicam	0.3-0.6 mg/kg	SC/PO	q24h	Excellent synergistic effect with opioids

Rabbit Formulary				
Carprofen	2-4 mg/kg 1.0-2.2 mg/kg ¹	SC PO	q24h q12h	Excellent synergistic effect with opioids
Anesthetics	Dose (mg/kg)	Route	Frequency	Comments
INHALATION				
Recommended: Isoflurane	3-5% induction 1-3% maintenance	Inhalation	continuous	Must use precision isoflurane vaporizer
INJECTABLE				
Recommended: Ketamine/ Xylazine	35-50 mg/kg 5-10 mg/kg	IM	If additional doses needed to prolong anesthesia provide 1/3 dose ketamine only	Minor surgery only; lower doses used for anesthetic induction when isoflurane used for anesthetic maintenance
Ketamine/ Medetomidine	15-25 mg/kg 0.25-0.5 mg/kg	SC/IM		Minor surgery only; used for anesthetic induction when isoflurane used for anesthetic maintenance
Ketamine/ Xylazine/ Buprenorphine	35 mg/kg 5 mg/kg 0.03 mg/kg	IM	If additional doses needed to prolong anesthesia provide 1/3 dose ketamine only	Minor surgery only; used for anesthetic induction when isoflurane used for anesthetic maintenance
Ketamine/ Medetomidine/ Buprenorphine	35 mg/kg 0.5 mg/kg 0.03 mg/kg	IM	If additional doses needed to prolong anesthesia provide 1/3 dose ketamine only	Minor surgery only; used for anesthetic induction when isoflurane used for anesthetic maintenance
Reversal agents	Dose (mg/kg)	Route	Frequency	Comments
Atipamezole	5X medetomidine (same volume as medetomidine)	IM preferred/ IV		More specific for reversal of medetomidine than xylazine
Yohimbine	0.2-1.0 mg/kg ¹	IV slowly/ IM	Once	Reverses sedative, cardiodepression, analgesic effects of xylazine
Local anesthetics	Dose (mg/kg)	Route	Frequency	Comments
Bupivacaine / Lidocaine mixture	1.5 mg/kg 0.5 mg/kg	SC	Once	Mix in same syringe for infiltration or local nerve block. Bupivacaine has slow onset (20-30 minutes), longer duration (4-8 hours); Lidocaine has rapid onset (5-10 minutes), shorter duration (1-2 hours).

References

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Other references:

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Figure 1. Mouse Grimace Scale⁴

Figure 1 | In the MGS, intensity of each feature is coded on a three-point scale. For each of the five features, images of mice exhibiting behavior corresponding to the three values are shown.
