

RAT AND MOUSE ANESTHESIA AND ANALGESIA
Formulary and General Drug Information

Approved: March 2nd, 2016

Definitions

General Anesthesia: Loss of consciousness in addition to loss of sensation

Analgesia: Loss of sensitivity to pain.

****Anesthesia does not necessarily equate with analgesia! ****

General anesthesia produces loss of consciousness, so the animal cannot consciously perceive pain, but in unconscious animals, painful stimuli will still be transmitted and processed by the central nervous system. Although the animal does not perceive pain during the surgery, central hypersensitivity can still develop in the spinal cord and brain causing perception of postoperative pain to be heightened. Some anesthetics, such as the alpha-2 adrenoreceptor agonists (i.e. Xylazine, Dexmedetomidine), do have some analgesic properties. In addition, additional analgesics can be used as part of the anesthetic regimen (i.e. opioids, non-steroidal anti-inflammatories).

Associated Guidelines and SOPs can be found on the ACC SOP/Guidelines website:

<http://ors.ubc.ca/contents/animal-care-sops-guidelines>

- Policy 16 Survival Surgery of Rodents
- SOP ACC-01-2015 Rodent Anesthesia
- SOP ACC-02-2014 Rodent Survival Surgery
- Surgical Class and Analgesia Guidelines
- TECH 17 Buprenorphine SOP (2014)
- TECH 19 Metacam SOP (2016)
- TECH 18 Ketoprofen SOP (2016)
- TECH 16 Local Anesthetic SOP (2014)

FORMULARY FOR MICE

Local anesthetic/analgesics			
Lidocaine hydrochloride (2%)	Dilute to 0.5%, do not exceed 7 mg/kg total dose, SC or intra-incisional	Use locally before making surgical incision	Faster onset than bupivacaine but short (<1 hour) duration of action
Bupivacaine (0.5%) (Marcaïne) (Recommended)	Dilute to 0.25%, do not exceed 8 mg/kg total dose, SC or intra-incisional	Use locally before making surgical incision	Slower onset than lidocaine but longer (~ 4-8 hour) duration of action
Ketamine combinations			
Ketamine-Dexedetomidine (Recommended)	K:75-150 mg/kg + D:~0.5-1 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedures. If redosing, use 1/3 dose of ketamine alone-may lose surgical anesthesia. Dexmedetomidine may be reversed with Atipamezole.	
Ketamine-Xylazine	K: 75-150 mg/kg + X: 16-20 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedure. If redosing, use 1/3 dose of ketamine alone-may lose surgical anesthesia. Xylazine may be reversed with Atipamezole or Yohimbine.	
Ketamine-Xylazine-Acepromazine (Recommended)	K:75-100 mg/kg + X:16-20 mg/kg +A: 3 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedures. If redosing, use 1/3 dose of ketamine alone. Xylazine may be partially with Atipamezole or Yohimbine.	
Ketamine-Midazolam	K: 75-100 mg/kg + M: 4-5 mg/kg IP or SQ (in same syringe)	Will not produce surgical-plane anesthesia for surgical procedures, but may be useful for restraint.	
Other injectable anesthetics			
Sodium pentobarbital (Nembutal)	50-90 mg/kg IP	Recommended for terminal/acute procedures only, with redosing as needed. May occasionally be appropriate for survival procedures. Dilute to 9.1 mg/ml for use (do not use the euthanasia solution).	Consider supplemental analgesia (opioid or NSAID) for invasive procedures, especially when used on a survival basis. For surgery, do not use the euthanasia solution which is typically 240 mg/ml
Opioid analgesia			
Buprenorphine (Recommended)	0.05-0.1 mg/kg SC	Used pre-operatively for preemptive analgesia and post-operatively every 8-12 hours	For major procedures, require more frequent dosing than 12 hour intervals. Consider multi-modal analgesia with a NSAID. High doses of buprenorphine may lead to pica behavior in rats.
Non-steroidal anti-inflammatory analgesia (NSAID) Note that prolonged use may cause renal, gastrointestinal, or other problems. Avoid using longer than 2 to 3 days.			
Carprofen	5 mg/kg SC or orally	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine.

Meloxicam (Recommended)	5 mg/kg SC or orally	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine. Recommended to give concurrent SQ fluids.
Ketoprofen (Recommended)	5 mg/kg SC	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine. Recommended to give concurrent SQ fluids.
Inhalation anesthetics			
Isoflurane (Recommended)	1-3% inhalant to effect (up to 5% for induction).	Survival surgery requires concurrent preemptive analgesia. Must use precision vaporizer	

Recommended dilutions for mice and small rats to get accurate volumes:

NOTE: some drugs are sensitive to light. Store diluted preparations away from light. Diluted drugs should be discarded 30 days after dilution.

Ketamine (100 mg/ml): Dilute to 50 mg/ml. Draw 1 ml of Ketamine (100 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 1 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Ketamine 50 mg/kg, the date prepared and the expiry date.

Dexmedetomidine (0.5 mg/kg): Dilute to 0.25 mg/kg. Draw 1 ml of Dexmedetomidine (0.5 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 1 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Dexmedetomidine 0.25 mg/kg, the date prepared and the expiry date.

Xylazine (20 mg/ml): Dilute to 2 mg/ml. Draw 1 ml of Xylazine (20 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 9 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Xylazine 2 mg/ml, the date prepared and the expiry date.

Acepromazine (10 mg/ml): Dilute to 1 mg/ml. Draw 1 ml of Acepromazine (10 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 9 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Acepromazine 1 mg/ml, the date prepared and the expiry date.

Midazolam (5 mg/ml): Dilute to 1 mg/ml. Draw 1 ml of Midazolam (5 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 4 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Midazolam 1 mg/ml, the date prepared and the expiry date.

Sodium Pentobarbital (54.7 mg/ml): Dilute to 9.1 mg/ml. Draw 1 ml of Sodium Pentobarbital (54.7 mg/ml) into a sterile syringe and dispense into a sterile multi-dose vial. To this, add 5 ml of sterile, pyrogen-free water or sterile 0.9% NaCl. Label the vial with Sodium Pentobarbital 9.1 mg/kg, the date prepared and the expiry date.

FORMULARY FOR RATS

Local anesthetic/analgesics			
Lidocaine hydrochloride (2%)	Dilute to 0.5%, do not exceed 7 mg/kg total dose, SC or intra-incisional	Use locally before making surgical incision	Faster onset than bupivacaine but short (<1 hour) duration of action
Bupivacaine (0.5%) (Marcaine) (Recommended)	Dilute to 0.25%, do not exceed 8 mg/kg total dose, SC or intra-incisional	Use locally before making surgical incision	Slower onset than lidocaine but longer (~ 4-8 hour) duration of action
Ketamine combinations			
Ketamine-Dexmedetomidine (Recommended)	K: 75-90 mg/kg + D: 0.5-0.75 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedures. If redosing, use 1/3 dose of ketamine alone-may lose surgical anesthesia. Dexmedetomidine may be reversed with Atipamezole.	
Ketamine-Xylazine	K: 75-90 mg/kg + X: 5-10 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedures, though more reliable than in mice. If redosing, use 1/3 dose of ketamine alone-may lose surgical anesthesia. Xylazine may be reversed with Atipamezole or Yohimbine.	
Ketamine-Xylazine-Acepromazine	K: 75-90 mg/kg + X: 5-10 mg/kg +A: 1-2 mg/kg IP or SQ (in same syringe)	May not produce surgical-plane anesthesia for major procedures. If redosing, use 1/3 dose of ketamine alone. Xylazine may be reversed with Atipamezole or Yohimbine.	
Ketamine-Midazolam	K: 75-90 mg/kg + M: 4-5 mg/kg IP or SQ (in same syringe)	Will not produce surgical-plane anesthesia for surgical procedures, but may be useful for restraint.	
Other injectable anesthetics			
Sodium pentobarbital	40 -60 mg/kg IP	Recommended for terminal/acute procedures only, with booster doses as needed. May occasionally be appropriate for survival procedures. Dilute to 9.1 mg/ml for use (do not use the euthanasia solution).	Consider supplemental analgesia (opioid or NSAID) for invasive procedures, especially when used on a survival basis. *For surgery, do not use the euthanasia solution which is typically 240 mg/ml
Opioid analgesia			
Buprenorphine (Recommended)	0.01 - 0.05 mg/kg SC or IP	Used pre-operatively for preemptive analgesia and post-operatively every <u>8</u> -12 hour	For major procedures, require more frequent dosing than 12 hour intervals. Consider multi-modal analgesia with a NSAID. High doses of buprenorphine may lead to pica behavior in rats.
Non-steroidal anti-inflammatory analgesia (NSAID) Note that prolonged use may cause renal, gastrointestinal, or other problems. <u>Avoid using for longer than 2 to 3 days.</u>			
Carprofen	5 mg/kg SC or orally	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days.	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine.

Meloxicam (Recommended)	1-2 mg/kg SC or orally	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days.	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine. Recommended to give concurrent SQ fluids.
Ketoprofen (Recommended)	5 mg/kg SC	Used pre-operatively for preemptive analgesia and post-operatively every 24 hours for 3 days.	Depending on the procedure, may be used as sole analgesic, or as multi-modal analgesia with buprenorphine. Recommended to give concurrent SQ fluids.
Inhalation anesthetics			
Isoflurane (Recommended)	1-3% inhalant to effect (up to 5% for induction).	Survival surgery requires concurrent preemptive analgesia. Must use precision vaporizer	

Reversal agents (Mice and Rats)			
Atipamezole	1-2.5 mg/kg SC, IP, or IV	For reversal of Medetomidine or Xylazine effects	More specific for medetomidine than for xylazine. Note reversal of alpha-2 agonist results in removal of analgesic properties so additional analgesia must be given prior to reversal if painful procedure has been performed.
Yohimbine	1.0 – 2.0 mg/kg SC, IP or IV	For reversal of xylazine effects	Note reversal of alpha-2 agonist results in removal of analgesic properties so additional analgesia must be given prior to reversal if painful procedure has been performed.

For purposes of administering a drug via the drinking water

	Rat	Mouse	Hamster	Gerbil	Guinea Pig
Normal* Daily Water Consumption	8-11 ml/100 gm body weight/day	15 ml/100 gm body weight/day	30 ml/day	4-7 ml/100gm body weight/day	10 ml/100 gm body weight/day

*Animals that have been subjected to a painful procedure/surgery will not drink the “normal” amount of water for a minimum of 24 hours post-surgery/post-procedure. It is estimated that normal water consumption will be reduced by at least 50%.

GENERAL INFORMATION ON COMMONLY USED ANESTHETICS AND ANALGESICS IN RODENTS

INHALANT AGENTS

Isoflurane, Sevoflurane and *Halothane

Advantages	Disadvantages
Ability to control depth and duration of anesthesia	Requires specialized equipment to administer - expensive
Fast, controlled induction and recovery from anesthesia	Potential for human exposure – must scavenge waste gases
Not a controlled substance	No analgesia provided
Minimal metabolism required so can use in old, sick, young, or animals with altered organ function	
The carrier gas, oxygen, helps provide support during the procedure	

The standard inhalant anesthetic for laboratory animal is isoflurane, delivered to effect in concentrations of 1-3% in oxygen (up to 5% for initial induction), using a precision vaporizer.

Isoflurane

- This is probably the most commonly used inhalant anesthetic in laboratory animals currently.
- It is a profound respiratory depressant at higher concentrations
- Has quick induction (3-5 minutes at 5%) and recovery times
- Is a respiratory irritant so animals will initially hold their breath or hold their heads up to avoid breathing it in
- It is heavier than air in its vapor form so will “sink” to the lowest portion of the area it is being used in.
- It reduces pain sensitivity but is not an analgesic (it simply causes unconsciousness)
- Causes muscle relaxation
- Minimal effects on heart rate and blood pressure at surgical levels of anesthesia
- Minimal (<1%) metabolism of isoflurane occurs

Sevoflurane

- Not yet commonly used in laboratory animals but will likely become more popular (current about 3 times the cost of isoflurane).
- Causes respiratory depression at higher concentrations (though less than isoflurane)
- Have very quick induction (less than 1 minute) and recovery times
- Does not irritate the respiratory tract so is “pleasant” to breathe (smells a bit like vanilla)
- Is associated with increased intracranial pressure (avoid in head trauma models)
- It reduces pain sensitivity but is not an analgesic (it simply causes unconsciousness)
- Causes muscle relaxation
- Minimal effects on heart rate and blood pressure at surgical levels of anesthesia
- Some metabolism (approximately 5%) of sevoflurane occurs

***Halothane** used to be very commonly used in both human and animal anesthesia but it has been removed from the market due to the health concerns of chronic exposure to the waste anesthetic (linked to miscarriages and liver cancer).

Occupational safety is a serious concern. Inhalants must be directly vented out of the room, or, less reliably, adsorbed in a charcoal canister filter. Filters must be weighed and replaced before they reach target weight (usually an increase of 50 gm). Note that charcoal filtration is not accepted as a safe scavenging system by UBC Health, Safety and Environment.

INJECTABLE ANESTHETICS

Advantages	Disadvantages
Typically inexpensive	Lack of ability to control depth or duration of anesthesia
No specialized equipment require to administer	Prolonged recovery – must metabolize and excrete the drug(s) in order to recovery
Easy to administer (SQ or IP injection)	Many of the drugs used are controlled substances so an exemption from Health Canada is required.
Low risk of human exposure	High risk of overdosing if repeated dosing required
May provide some analgesia depending on the drug chosen	Higher risk in very young or older animals with poorer organ function
	Profound physiological effects
	Significant differences in response of individuals to the same dose

Ketamine (i.e. Ketalean, Ketaset)

Ketamine is a dissociative anesthetic used in a wide variety of species. In low doses, ketamine provides chemical restraint with minimal analgesia. In most instances, ketamine is used in combination with other injectable agents.

- Is a dissociative general anesthetic (patient is not fully unconscious)
- Inhibits NMDA-receptors.
- Is a controlled substance (due to potential of abuse by humans) so requires an exemption from Health Canada
- Causes muscle rigidity and partial immobilization (no relaxation)
- Used on its own will not induce a surgical depth of anesthesia (*typically used in combination with either xylazine or dexmedetomidine to get surgical plane of anesthesia)
- Little to no analgesic properties at the doses typically used
- Can lower the threshold for seizures so caution should be used when using in animals prone to seizures, brain trauma or increased intracranial pressure
- Has a high therapeutic index so the dose range is quite wide
- Has less effect on the heart rate and blood pressure
- Can be associated with increased intraocular pressure (usually not a concern unless the animal already has increased intraocular pressure or the surgery involves the inside of the eye).
- Can be associated with excitement upon recovery (animals may become “jumpy” and stumble around the cage and may injure themselves until they are fully recovered)
- Is acidic so injections are painful (“sting”) – avoid intramuscular injections of this drug.

Xylazine (i.e. Rompun) and *Dexmedetomidine (i.e. Dexdomitor)

- *Dexmedetomidine is the newer, more “pure” version of Medetomidine and the doses are different
- Are sedatives so used on their own will not cause unconsciousness at safe dosage levels
- Are alpha-2 adrenergic receptor agonists
- Are not yet a controlled substance so easier to purchase (though this may change since it is becoming more popular as a drug of abuse)
- Cause muscle relaxation
- Used on their own, will not induce a surgical depth of anesthesia (*either are typically used in combination with ketamine to get surgical plane of anesthesia).
- Some analgesic properties (but for moderate to severe pain, additional analgesia must be used)
- Have a low therapeutic index so the dose range is quite narrow
- Have significant effect on heart rate and blood pressure (decreases both heart rate and blood pressure)
- Are associated with decreased peripheral blood flow but increased blood flow through major internal organs so acts as a diuretic (to cause increased urine production) and this can lead to dehydration
- Cause respiratory depression
- Cause transient decrease of tissue sensitivity to insulin so results in hyperglycemia (this can last for many hours)
- Cause piloerection (so heat support is important) and exophthalmos (“bulging” eye) so protecting the cornea from damage and drying out is important
- Can be reversed using reversal agents – this speeds recovery and reverses the negative side effects but also reverses the analgesia effects of the drugs.

Diazepam (i.e. Valium) and Midazolam (i.e. Versed)

- Diazepam is fat soluble (slower absorption when given SQ or IP) and Midazolam is water soluble (faster absorption when given SQ or IP)
- Are mild sedatives so used on their own will not cause unconsciousness but do calm and relax the animal
- Can be combined with Ketamine to perform non-invasive, non-painful procedures such as imaging
- Are benzodiazepines
- Are controlled substances (due to potential of abuse by humans) so requires an exemption from Health Canada
- Cause muscle relaxation and will reduce the doses of other anesthetic drugs when used in combination with other anesthetic drugs
- Have no analgesic properties
- Have anti-seizure properties
- Have higher therapeutic index
- Cause minimal cardiovascular and respiratory depression

Ketamine combinations

Ketamine and alpha2-agonists (Xylazine or Dexmedetomidine)

Ketamine may be combined with the alpha-2 agonists, Xylazine or dexmedetomidine, in the same syringe mixed right before injecting, to produce a deep level of sedation. In some situations and in some species and strains, an adequate depth of anesthesia for surgery may be attained. In other cases, this sedation may require an inhalant agent to achieve surgical anesthesia. It is generally safer to titrate to effect with inhalant anesthetic from a precision vaporizer than with supplemental injections of ketamine.

Advantages: Advantages of ketamine/alpha2-agonist combinations are that they may be combined in one syringe, that they may produce short-term surgical anesthesia with good analgesia, and that recovery can be hastened by reversing the alpha2-agonist with Atipamezole or Yohimbine.

Disadvantages: Disadvantages of ketamine/alpha2-agonist combinations are that they will not reliably reach surgical plane of anesthesia in all cases, and that they can cause profound cardiac depression. The reversal of the alpha2-agonist results in the reversal of the analgesic component of the combination and should only be done when there is concern for the animal.

Caution for use: If a ketamine/alpha2-agonist combination is used for surgery longer than 20 minutes, animals will likely require additional anesthetic. Redosing with a lower dose of ketamine rather than the combination is usually safer, as the cardiovascular depression of alpha2-agonists is often longer-lasting than the sedation or analgesia produced. However, redosing repeated with ketamine alone will not produce a surgical plane of anesthesia. Preferably, an inhalant anesthetic should be used to continue the anesthetic for procedures longer than 30 minutes.

These combinations should be mixed in the syringe immediately prior to use since the drugs are incompatible and their efficacy, once mixed, will decrease over time.

Ketamine and Benzodiazepines (Midazolam or Diazepam)

Ketamine may also be combined with the benzodiazepines, Midazolam or Diazepam, in the same syringe to produce a deep level of sedation. In most cases, this sedation will require an inhalant agent or other anesthetic to achieve surgical anesthesia. In most applications, Midazolam is preferred, as it can be injected intramuscularly; intramuscular injection of propylene glycol (the carrier in injectable diazepam) can cause painful, sterile abscesses and is discouraged.

Advantages: Advantages of ketamine/benzodiazepine combinations are that they may be combined in one syringe, mixed immediately prior to injection, are partially reversible and will produce deep sedation.

Disadvantages: Disadvantages of ketamine/benzodiazepine combinations are that they will not reliably reach surgical anesthesia. This combination, however, is preferred for imaging and other non-painful

procedures as it is safer than the ketamine/alpha2-agonist combinations. Diazepam should be restricted to intravenous or intraperitoneal use. Pharmacologically, Telazol is a dissociate-benzodiazepine combination. These drugs are also controlled and require a license for use.

These combinations should be mixed immediately before use and protected from light. Benzodiazepines in mice may cause increased salivation and should be pre-treated with an anticholinergic (such as atropine or glycopyrrolate).

Barbiturates (Sodium Pentobarbital – i.e. Somnotal, Nembutal)

Though superseded in most applications by newer anesthetics, barbiturates still have their place in the research animal laboratory. They are most frequently used in terminal or acute studies, as recovery can be prolonged and unpleasant. Barbiturates are often the anesthetic of choice when neurophysiological recordings such as visual or auditory evoked responses are being conducted. Concurrent use of an analgesic (opioid or non-steroidal anti-inflammatory drug) is encouraged as it may improve pain relief with barbiturate use, and lower the required dose of barbiturate.

Sodium pentobarbital (**Nembutal**) and sodium thiopental (**Pentothal**) are currently the two most commonly used barbiturates. The duration of action of pentobarbital is considerably longer than that of thiopental.

- Can be a sedative, anesthetic agent or euthanasia agent depending on the dose
- Is a barbiturate
- Is a controlled substance (due to potential of abuse by humans) so requires an exemption from Health Canada
- Has no analgesic properties
- Has a low therapeutic index so the dose range is quite narrow
- Causes dose dependent depression of heart rate, blood pressure and respiration
- Is acidic so will cause pain when injected – diluting the drug in sterile water prior to injecting is recommended (dilute to approximately 9 mg/ml)
- Has significant individual and strain differences in response to the same dose and males are known to be more sensitive than females.

Non-Recovery Anesthetics

****These drugs are approved for only non-recovery procedures in which the animal will not be permitted to recover from the anesthetic due to the severe physiological changes these drugs cause in animals. The use of these must be justified on the animal care protocol with an explanation why no other anesthetic drug can be used.**

Chloral hydrate and Alpha-chloralose

- Both are considered hypnotic agents rather than anesthetic agents
- Chloral hydrate lasts 1-2 hours while alpha-chloralose lasts 8-10 hours
- Both have poor analgesic properties

- Have little effect on the cardiovascular or respiratory system at hypnotic doses (but these doses do not induce anesthesia) but larger doses cause severe respiratory depression, severe decrease in blood pressure and cardiac arrhythmias
- Are mutagenic
- Cause severe inflammation of the peritoneal cavity when injected intraperitoneally (causes peritonitis) and also cause the intestines to stop moving (ileus) which can be fatal. Can also cause gastric ulcers in rodents.

Urethane

- Long-lasting anesthetic (8-10 hours)
- Has minimal cardiovascular depression
- Carcinogenic and mutagenic in animals and people so proper safety precautions must be taken when handling this drug in both the powder and liquid form – it well absorbed across the skin and through lungs and mucous membranes (suppresses bone marrow, crosses the placenta, induces tumor formation and can initiate pre-neoplastic changes in the skin)
- Must also take safety precautions when handling tissues and blood from animals anesthetized with urethane

Tribromoethanol (Avertin)

- Short acting (15 minutes of surgical anesthesia)
- Not available commercially so must be prepared and drug deteriorates over time (must be stored in the dark and refrigerated)
- Causes intestinal ileus (intestine stops moving), muscle necrosis, and peritonitis

Analgesics

Opioids

Opioid drugs are important components of many surgical anesthesia regimens, and are the most potent available post-procedural analgesics. Drugs in this group vary in their potency as well as their duration of action. Fentanyl, oxymorphone, buprenorphine and butorphanol are the most commonly used opioids in laboratory animal care, though others may be used on occasion. Fentanyl is the most potent of the three, but also the shortest acting. Buprenorphine is the longest acting and is good for most post-operative applications. Buprenorphine and butorphanol are mixed agonist/antagonists at different opioid receptors; they produce a less profound respiratory depression than full agonists, but also have a “ceiling effect” in the degree of analgesia produced with increasing doses.

Opioids are most often administered by injection. Oral use can be effective, but requires much higher doses because of “first-pass” liver metabolism when absorbed from the gut.

Pre-emptive analgesic involves using appropriate analgesics before they are needed (i.e. prophylactically) and is strongly recommended as it will prevent pain “windup”, decreasing the requirement in the post-operative period -- buprenorphine may be administered when the general anesthetic is administered, or at any time during surgery. Respiratory depression is minimal, though

sleep time may be lengthened. Pre-emptive use enhances pain management during the immediate post-surgical period. Though it increases animal handling (a stressor), administration of the analgesic 30 minutes prior to the initial surgical incision maximizes the analgesic efficacy in most situations.

Advantages: Opioids are potent analgesics. Concurrent use with inhalant or injectable general anesthesia will lower the required dose of the anesthetic to maintain a surgical plane of anesthesia.

Disadvantages: Opioids can suppress respiration (more marked effect with the use of fentanyl than with buprenorphine). Opioids may increase locomotor activity, and may cause pica (abnormal ingestion of non-food items such as bedding) in rats. Alternatively, they may sometimes cause sleepiness and slower recovery from general anesthesia. Opioids are controlled substances and require an exemption license from Health Canada for use.

Cautions for use: Buprenorphine has found favor as the longest-acting opioid analgesic with the least effect on immune function. However, this duration of action is closer to 6 hours in most situations than it is to 12 hours. Twelve hours is the absolute maximum dosing interval for use of buprenorphine for post-procedural pain in rodents.

****See Use of Buprenorphine in Rodents SOP TECH 17**

Non-steroidal anti-inflammatory drugs (NSAIDs)

The advent of newer, more potent, more specific anti-inflammatory agents has increased their usefulness in laboratory animal use. Most reduce fever, reduce inflammation, and provide varying degrees of analgesia (acetaminophen does not significantly reduce inflammation).

Advantages: Carprofen, ketoprofen, and meloxicam may have duration of analgesic action up to 24 hours. They may be used concurrently with anesthetics, with opioid analgesics, and with local anesthetic/analgesics. Injectable NSAIDs are useful for accurate dosage and administration to small rodents. They are not controlled substances (some are by veterinary prescription only) and must be obtained through the Animal Care Centre.

Disadvantages: NSAIDs may decrease clotting ability, of possible concern following surgery. Gastric upset and even ulceration may occur, especially with prolonged use. Prolonged use carries the risk of kidney or liver disease. Undesired side effects are more likely with increasing length of usage -- for most situations, limit use of NSAIDs to 3 days per animal, except under veterinary supervision. Do not use in dehydrated animals or in animals with kidney or liver dysfunction. It is recommended to give 10 ml/kg of sterile Lactated Ringer's Solution or 0.9% saline SQ at the same time as the NSAID to ensure good hydration.

****See Use of Ketoprofen in Rodents SOP TECH18**

****See Use of Meloxicam in Rodents SOP TECH19**

Local anesthetic/analgesic drugs (lidocaine and bupivacaine)

Local anesthetic/analgesic drugs (lidocaine and bupivacaine) may be useful both during surgery, and post-operatively. They block nerve conduction when applied locally at sufficient concentration. In larger animals, Lidocaine has a fast onset of action and provides a couple of hours of analgesia (less than 1 hour in rodents). Bupivacaine has a slower onset of action (up to 30 minutes) but provides up to 8-12 hours of residual analgesia in some animals (closer to 8 hours in rodents). Both of these drugs are thought to provide less duration of analgesia in rodents. Both can be infiltrated subcutaneously at the surgical site. Lidocaine cream (EMLA) is used topically on shaved, intact skin prior to venipuncture, though it requires 30-60 minutes or more of contact with skin to reach full effect. It is not a substitute for injectable local anesthetics for surgical procedures as it only penetrates the skin and is often removed by the animal before it can have an effect.

Even with general anesthesia, the topical or subcutaneous (at surgical incision site) administration of a local anesthetic is recommended in order to provide additional post-surgical analgesia. Local anesthetics should not be used alone to provide post-surgical/post-procedural analgesia.

Advantages: Intra-operative use can augment the pain relief of general anesthetics and reduce the need for frequent redosing of opioids and/or NSAIDs. They are not controlled substances. At appropriate doses, they have minimal cardiovascular effect.

Disadvantages: Intramuscular and intravenous injection should both be avoided. Systemic toxicity (including seizures and death) can result from over dosage (more likely to occur with smaller animals) and with accidental intravenous injection. Lidocaine may sting when first injected.

****See Use of Local Anesthetics in Rodents SOP TECH16**

SPECIES-SPECIFIC CONSIDERATIONS

In general, smaller animals have higher metabolic rates and frequently require higher doses of anesthetics and analgesics at more frequent intervals to achieve the desired effect. Species, strain and age differences often overshadow this general principle however. It is always best to start with a drug regimen developed in the species, age and strain with which the Principal Investigator is working, rather than extrapolate from one species or strain to another.

Mice and Rats

Isoflurane is recommended as the first choice anesthetic in mice and rats. It should be delivered at a known percentage (1-3% for maintenance; up to 5% for induction) in oxygen from a precision vaporizer.

Anesthetic monitoring of small rodents includes testing of rear foot reflexes *before* any incision is made, and continual observation of respiratory pattern, mucous membrane color, and responsiveness

to manipulations and rear foot reflexes throughout the procedure. Rectal temperature and heart rate should be monitored electronically during long or involved procedures.

Injectable anesthetics are typically administered by intraperitoneal or subcutaneous route. Injectable analgesics and reversal agents are often administered by the subcutaneous or intravenous route. Intramuscular injections must generally be avoided because of the small muscle mass. Diluting drugs in sterile water or saline solution is necessary to accurately measure volume for injection for mice or small rats. It may also make some drugs less irritating when injected. Dilution may decrease shelf-life; the standard is to discard drugs within 30 days of dilution. Vials containing sterile, diluted drugs must be labeled with the contents, concentration and the expiration date.

Ketamine/alpha-2 agonist combinations produce short-duration surgical anesthesia in larger species, but are frequently insufficient for major surgical procedures in many strains of mice and some rats. An excellent approach is to use a ketamine combination, but then titrate to effect with isoflurane from a precision vaporizer. Safety and efficacy should be demonstrated in a pilot group of animals before a large-scale study is initiated. Partial reversal of the xylazine or dexmedetomidine using yohimbine or atipamezole is possible, and will restore cardiovascular status more quickly but will reverse the analgesic properties of these drugs so additional analgesia must be given if a painful procedure was performed.

Mice and rats are nocturnal animals and are frequently housed in groups of nearly identical animals. These two factors make diagnosis of mild to moderate pain challenging. Weight loss is an important parameter to monitor in animals at risk for on-going pain. Pre-emptive treatment of pain, before signs of pain are apparent, is recommended.

Isoflurane provides no post-operative pain relief. If used for surgery, concurrent and follow-up use of buprenorphine and/or a non-steroidal anti-inflammatory will be necessary. The veterinary staff recommends injecting the analgesic 30 minutes prior to the *start* of surgery.