

December 2, 2009

## ANIMAL CARE AND USE PROGRAM

### Pain, Distress, Analgesia, and Anesthesia

1. **POLICY:** Research investigators will adhere to all animal care and use guidelines to reduce or eliminate discomfort, distress and pain to animal subjects. The relief of pain and distress is ethically sound, humane, and promotes good science.

#### 2. **PROCEDURES:**

a. **RESEARCH PROPOSAL:** Each research proposal involving animals must indicate all details of animal use. All aspects that may use more than momentary pain and/or distress must be addressed; alternatives to painful or distressful procedures must be considered; and methods, anesthetics and analgesics to minimize or eliminate pain and distress must be included for any painful or distressful procedure that cannot be relieved or minimized. Only the exceptions that are justified in the Animal Component of the research proposal and approved by the Institutional Animal Care and Use Committee (IACUC) are permitted.

b. **RECOGNIZING PAIN OR DISTRESS, MORBIDITY:** Animals must be monitored for evidence of pain or distress. Critical to the assessment of the presence of pain/distress is the ability to distinguish between normal and abnormal animals behavior. Attachment 1 identifies signs associated with animal pain or distress..

c. **SEDATIVES, TRANQUILIZERS, ANALGESIA, AND ANESTHESIA:** The attached species-specific information on sedatives, tranquilizers, analgesia, and anesthesia provides information for investigator use in preparing the research proposal (attachment 2). The Principal Investigator should consult with the veterinarian prior to submitting the research proposal.

d. **WITHHOLDING OF SEDATIVES, ANALGESICS, OR ANESTHETICS:** In experiments that cause pain or distress, the withholding of sedatives, analgesics, or anesthetics must be justified scientifically by the Principal Investigator in writing and approved by the IACUC.

e. **GUIDANCE:** All procedures that cause more than momentary/slight pain or distress, and the use of anesthetics and analgesics must be discussed with the veterinarian in the planning stage prior to submission of the animal protocol for review. Subsequent changes should be brought to the attention of the veterinarian by the investigator..

f. **MONITORING.** The VMU supervisor monitors usage by means of monthly checks of drugs dispensed through the VMU. The investigators utilizing surgical studies in the VMU maintain anesthetic records. The VMU supervisor will make periodic checks of the research technician's data. Random biannual checks are made by the IACUC facility inspection team.

g. **TRAINING AND EXPERIENCE:** Research personnel caring for and using animals must receive training in, and be responsible for, assessing pain and distress in the animals. VMU animal technicians and research technicians performing anesthesia must have experience in the techniques used. Training is obtained through on-the-job training and continuing education. The VMU supervisor will certify the

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competency of VMU personnel and the principal investigators will certify same competency of their personnel conducting procedures.

h. **VOLATILE ANESTHETICS:** The use of explosive anesthetics is not permitted within the Veterinary Medical Unit. A waste gas scavenging mechanism is incorporated in the inhalation anesthesia apparatus in the OR suite and treatment room R222.

i. **MORIBUND CONDITIONS:** Attachment 3 provides the investigator signs and symptoms for judging moribund conditions (state of dying) in rodents. The investigator and his staff have primary responsibility for proper use and monitoring. The veterinarian, VMU supervisor and VMU staff provide additional oversight. Rodents in Class C studies are monitored daily, including weekends and holidays.

3. **RESCISSION:** Research Service Memorandum 06-12, dated September 8, 2006.



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ACOS for Research and Development

### Attachments

1. Pain/Distress Symptoms
2. Pain/Distress Relievers
3. Moribund Symptoms

Distribution: Research Investigators

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**Attachment 1**

**POTENTIAL SIGNS ASSOCIATED WITH PAIN AN DISTRESS IN RODENTS AND RABBITS**

	Mice	Rats	Rabbits
Decreased Food and Water Consumption	X	X	X
Weight Loss	X	X	X
Self-imposed isolation/hiding	X	X	X
Self-mutilation, gnawing at limbs	X	X	X
Rapid breathing	X	X	X
Opened-Mouth Breathing	X	X	X
Abdominal Breathing	X	X	X
Grinding Teeth		X	X
Biting/Growling/Aggression		X	X
Increased/decreased movement	X	X	X
Unkempt Appearance (Erected, matted, or dull hair coat	X	X	X
Abnormal Posture/Positioning (e.g., head-pressing, hunched back	X	X	X
Restless Sleep			X
Tearing (including porphyria), Lack of blinking reflex		X	X
Dilated pupils			X
Muscle rigidity, lack of muscle tone	X	X	X
Dehydration/skin tenting/sunken eyes	X	X	X
Twitching, trembling, tremor	X	X	X
Vocalization (Rare)	X	X	X
Redness of Swelling Around Surgical Site	X	X	X
Increased Salivation			X

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### Attachment 2

#### SPECIES INFORMATION

MOUSE (*Mus musculus*)

Physiologic parameters:

Body temperature = 36.5-38.0°C

Heart rate = 325-780/min

Respiratory rate = 94-163/min

Tidal volume = 0.09-0.23 ml

The use of chloroform as an anesthetic agent is discouraged. Chloroform can cause renal tubular calcification and/or necrosis, particularly in male mice; DBA/2 strain most susceptible.

Avertin is made by mixing equal amounts of tribromyl ethyl alcohol and tertiary amyl alcohol (2.5% dilution). If Avertin is improperly prepared or stored in the light, it will break down into dibromoacetic acid and hydrobromic acid which can be lethal in 24 hours. **Freshly mixed solutions are strongly recommended for safe use.** The solution can be kept as long as 4 months if it is stored in the dark at 4 degrees C. The solution should be tested to ensure that it has a pH>5.

- \* The therapeutic dose for carbon dioxide is close to the lethal dose; very short acting. Concurrent administration of 10-50% O<sub>2</sub> is recommended.
- \*\* Best for minor surgery procedures only.

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**MOUSE (*Mus musculus*)**

<u>Drug indication and Drugs</u>	<u>Dosage and Route of Administration</u>	
<b>Restraint/Preanesthesia</b>		
Atropine	0.02-0.05 mg/kg	IM
Diazepam, C-IV (Valium®)	5 mg/kg	IP
Ketamine, C-III (Ketaset®, Vetalar®)	22-44 mg/kg	IM
Telazol®, C-III (for restraint)	100-160 mg/kg	IM IP
Carbon dioxide* + 10-50% O <sub>2</sub>	To effect	IH
<b>Anesthesia</b>		
Sodium Pentobarbital, C-II	50-90 mg/kg	IP
Ketamine**, C-III	50-200 mg/kg	IP
	40-60 mg/kg	IM
Avertin (Tribromoethanol)	125/250 mg/kg	IP
	0.02 ml/g (1.2% solution)	
<b>Ketamine/Kylazine:</b>		
- Add 7 mg xylazine to 35 mg Ketamine (dose based on ketamine)	70-80 mg/kg	IM IP
- Add 1.0 ml xylazine (100 mg/ml) and 1.0 ml ketamine (100 mg/ml) and 4.6 ml sterile water	0.1 ml/20 g	IM IP
Halothane (Fluothane®)	To effect	IH
Isoflurane	To effect	IH
<b>Analgesia</b>		
Morphine, C-II	5-10 mg/kg q2-4h	SC IP
Butorphanol tartrate (Torbugesic®), C-IV	2.5-5 mg/kg q1-2h	SC
Buprenorphine, C-V	2 mg/kg q12h	SC IP
Oxymorphone, C-II	0.15 mg/kg q4h	IM
Ketorolac	0.7-10 mg/kg q24h	PO

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### SPECIES INFORMATION

#### RAT (*Rattus norvegicus*)

Physiologic parameters:

Body temperature = 35.9-37.5°C

Heart rate = 250-450/min

Respiratory rate = 70-115/min

Tidal volume = 0.6-2.0 ml

Male rats and animals receiving low calorie diets require higher doses of barbiturates

Avertin has been reported to cause ileus in rats

The therapeutic dose for carbon dioxide is close to the lethal dose; very short acting. Concurrent administration of 10-50% O<sub>2</sub> is recommended.

The reversal agent, yohimbine, is only effective when xylazine or medetomidine has been used.

\* The projected duration of action for an analgesic is an approximation because the nature of the procedure and the level of pain that is experienced affect it.

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### RAT (*Rattus norvegicus*)

<u>Drug indication and Drugs</u>	<u>Dosage and Route of Administration</u>	
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#### Restraint/Preanesthesia

Atropine	0.04-0.1 mg/kg	SC
Diazepam, C-IV (Valium®)	0.5-15 mg/kg	IP
Ketamine, C-III (Ketaset®, Vetalar®)	22-50 mg/kg	IM
Carbon dioxide + 10-50% O <sub>2</sub>	To effect	IH

#### Anesthesia

Sodium Pentobarbital, C-II	30-60 mg/kg	IV IP
Ketamine, C-III (10 mg/ml solution)	50-90 mg/kg	IM
	50-100 mg/kg	IP
Ketamine/Xylazine:		
Ketamine	40-80 mg/kg	IM IP
Xylazine	10 mg/kg	IM IP
Halothane (Fluothane®)	To effect	IH
Isoflurane	To effect	IH
Carbon dioxide	To effect	IH
Telazol®, C-III	20-40 mg/kg	IIP
Ketamine/Medetomidine		
Ketamine	60-75 mg/kg	IP
Medetomidine (Domitor®)	0.25-0.5 mg/kg	SC
Chloral hydrate	(5% solution)	

#### Analgesia\*

Morphine, C-II	1.5-6 mg/kg q2-4h	SC
Butorphanol tartrate, C-IV (Torbugesic®)	2.5-5 mg/kg q1-2h	SC
Carprofen	5 mg/kg q12h	SC
Ketorolac	3-5 mg/kg q12-24h	PO
	1 mg/kg q12-24h	IM
Buprenorphine, C-V	0.01-0.05 mg/kg	SC IP

#### Reversal Agents

Yohimbine (reverses xylazine)	1-2 mg/kg	IM IP
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### SPECIES INFORMATION

#### HAMSTER (*Mescocricetus auratus*)

##### Physiologic parameters:

Body temperature = 37-38°C  
Heart rate = 250-500/min  
Respiratory rate = 35-135/min  
Tidal volume = 0.6-1.4 ml

Syrian or golden hamster is very resistant to morphine – no sedation or hypnotic effects

Syrian or golden hamster has an increased tolerance to pentobarbital.

#### HAMSTER (*Mesocricetus auratus*)

##### Drug indication and Drugs Dosage and Route of Administration

##### Restraint/Preanesthesia

Atropine	0.1 mg/kg	IP IM SC
Ketamine, C-III (Ketaset®, Vetalar®)	22-44 mg/kg	IM

##### Anesthesia

Sodium Pentobarbital, C-II	30-90 mg/kg	IP
Ketamine/Xylazine:		
Xylazine	10 mg/kg	IP IM
Ketamine	100 mg/kg	IP
Telazol®, C-III	20-80 mg/kg	IP IM
Halothane (Fluothane®)	To effect	IH
Isoflurane	To effect	IH
Ketamine/Diazepam, C-IV	0.5 mg/kg	IM IP

##### Analgesia

Buprenorphine, C-V	0.05-0.1 mg/kg q8-12h	SC IM
Butorphanol tartrate, C-IV (Torbugesic®)	1-5 mg/kg q2-4h	SC IM

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### SPECIES INFORMATION

#### RABBIT (*Oryctolagus cuniculus*)

Physiologic parameters:

Body temperature = 38-39.6°C

Heart rate = 130-325/min

Respiratory rate = 32-60/min

Tidal volume = 4-6 ml/kg

Many rabbits have serum atrophinesterase which causes reduced response to atrophine. Glycopyrrolate, another anticholinergic, can be used instead of atrophine.

Unique hypnotism or immobilization reflex has been observed in rabbits in the absence of drug use.

Large cecum can act as reservoir for anesthetics. Depending on drug solubility, the cecum can alter the pharmacologic effect.

Induction of anesthesia using volatile anesthetics (e.g., halothane and Isoflurane) should be done with caution due to initial breath holding when animals are first exposed to irritating gas vapors.

Give IV injections via marginal ear veins.

Self mutilation has been reported in rabbits after IM Ketamine administration. Dilution of ketamine with saline will limit this side effect.

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**RABBIT (*Oryctolagus cuniculus*)**

Drug indications and Drugs Dosage and Route of Administration

Restraint/Preanesthesia

Ketamine, C-III (Ketaset®, Vetalar®)	15-50 mg/kg	IM
Acetylpromazine	1.0-10 mg/kg	IM SC IV
Ketamine Acetylpromazine (10:1)	15-50 mg/kg	IM
Diazepam, C-IV (Valium®)	5-10 mg/kg	IV IM
Glycopyrrolate	0.005-0.011 mg/kg	IM
Butorphanol & Acepromazine		
Butorphanol tartrate, C-IV (Torbugesic®)	1 mg/kg	SC
Acetylpromazine	1 mg/kg	SC

Anesthesia

Sodium Pentobarbital, C-II (3% solution given slowly to effect)	15-40 mg/kg	IV
Ketamine/Xylazine/Acepromazine:		
Xylazine	5-10 mg/kg	IM
Ketamine, C-III	35-50 mg/kg	IM
Acepromazine	0.75 mg/kg	IM
Ketamine/Midazolam		
Ketamine, C-III	25 mg/kg	IM
Midazolam, C-IV	1 mg/kg	IM
Ketamine/Diazepam		
Ketamine, C-III	15-50 mg/kg	IM
Diazepam, C-IV	5-10 mg/kg	IM
Ketamine/Acepromazine/Butorphanol		
Ketamine, C-III	35 mg/kg	SC
Acepromazine	0.75 mg/kg	SC
Butorphanol tartrate, C-IV (Torbugesic®)	0.1 mg/kg	SC
Halothane (Fluothane®)	To effect	IH
Isoflurane	To effect	IH

Analgesia

Morphine, C-II	5 mg/kg q2-4h	SC IM
Acetylsalicytic Acid (Aspirin)	500 mg/kg	PO
Buprenorphine, C-V	0.02-0.1 mg/kg q8-12h	SC
Butorphanol tartrate, C-IV (Torbugesic®)	0.1-1.5 mg/kg q4h	IV
	1.0-7.5 mg/kg q4h	IM SC
Flunixin meglumine (Banamine®)	1.1 mg/kg q12h	IM SC
Carprofen	1.5 mg/kg q12h	PO
Ketoprofen	3 mg/kg q12h	IM

Reveral Agents

Yohimbine (reverses xylazine)

0.2 mg/kg

IV

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**Attachment 3**

**Signs and Symptoms for Judging the Moribund Condition (state of dying) in Rodents**

Signs and symptoms for morbidity plus:

1. Impaired ambulation (unable to reach food or water easily).
2. Evidence of muscle atrophy or other signs of emaciation (body weight is not always appropriate).
3. Extensive ulcerative dermatitis and infected tumors
4. Any obvious illness including such signs as lethargy (drowsiness, aversion to activity, physical or mental alertness), anorexia (loss of appetite, especially when prolonged), bleeding, difficult breathing, CNS disturbance, and chronic diarrhea.