Albert Einstein College of Medicine
Institute for Animal Studies
Van Etten 460
(718) 839-7100

Recommended Methods of Anesthesia, Analgesia, and Euthanasia for Laboratory Animal Species

Note: The following list has been compiled from a variety of sources. The Table includes Medications and Dosages currently recommended for each species. Each Species section contains IAS recommendations for Sedation, Anesthesia, Analgesia & Euthanasia. The species listed in the table are those currently included in IACUC Animal Use Protocols at Albert Einstein College of Medicine.

Doses may vary with species, strain, and other variables. All researchers are advised to consult with the IAS veterinary staff prior to initiating studies requiring surgical anesthesia and analgesia to determine proper starting doses. Subsequent, follow-up consultation is essential if any problems with anesthesia or alleviation of pain (analgesia) are encountered.

This list does not cover all species and certainly does not cover all effective agents or methods; but provides a basis for reference and discussion with the IAS veterinary staff. Please consult one of the references listed at the end of this document or one of the Institute for Animal Studies veterinarians if this list does not meet your needs.

MICE			
	Drug(s) or Method	**Dose (mg/kg)**	**Route**
ANTICHOLINERGICS	Atropine	0.02-0.05	SC,IM
	Glycopyrrolate	0.5	SC, IM
SEDATIVES/ IMMOBILIZATION	Diazepam	5	IP
	Ketamine / (+/- Acetylpromazine)	44-100 / 2-5	IP
	Xylazine	8 – 10	IP
	Ketamine	80-100	IP
INJECTABLE ANESTHESIA	Ketamine / Xylazine	80-100/10	IP
	Ketamine-Xylazine Mix [(1)]	0.1 ml/10g BW	IP
	Ketamine / Xylazine / Acetylpromazine	80-100 / 8-10 / 3	IP
	Ketamine-Xylazine-Acetylpromazine Mix [(2)]	0.1 ml/10g BW	IP
	Avertin (Tribromoethanol)[(3)]	240	IP
	Alpha-Chloralose[(4)](Non-Survival)	100-120	IP
	Urethane[(5)] (Non-Survival)	1000	IP
INHALANT ANESTHESIA	isoflurane	1-4%	inhalant
	ether[(6)]	to effect	inhalant
Analgesia	Flunixin (Banamine)	2.5	**SC**
	Ketoprofen	5	SC
	Meloxicam	5	SC
	Butorphanol (q 4 hr)	1 - 2	SC
	Buprenorphine (q 12 hr)	0.05 -0.2	SC
	Acetaminophen	200	PO

Euthanasia			
ACCEPTABLE METHODS	Carbon Dioxide	(Air Displacement Rate 10-30%/min)	Inhalant
	Pentobarbital	> 120	IV,IP
	Inhalant Anesthetic Overdose	to effect	inhalant
Conditionally [(8)] Acceptable Euthanasia Methods	Injectable Anesthetic Overdose	3x anesthesia dose	IV,IP
	Cervical Dislocation	Only if training documented	
	Decapitation	Only if training documented	
	Exsanguination (Under Anesthesia)	Non-Survival Surgery	

Abbreviations:
IM = intramuscular
IV = intravenous
BW = body weight
SC = subcutaneous
IP = intraperitoneal
q = every

FOOTNOTES

(1) KETAMINE-XYLAZINE COCKTAIL-MOUSE

The following regimen will produce a surgical level of anesthesia for 15-30 minutes and sedation of 1-2 hours.

Combine: [1.0ml] Ketamine (concentration: 100mg/ml)
[0.5ml] Xylazine (concentration: 20mg/ml)
ADD 8.5ml Normal saline 0.9% or PBS (Use Sterile Solution & Store in Sterile Container.

Mixtures of ketamine and xylazine are not stable and may lose potency. Make FRESH Mixture WEEKLY. Date of mixing should be placed on container or bottle used.

DOSE: 0.1ml per 10 gm of Body Weight
Delivered Dose is Ketamine 100 mg/kg **/** Xylazine 10mg/kg
Route:Intraperitoneal injection using 1 ml syringe 23-25 gauge 5/8 inch needle

(2) KETAMINE-XYLAZINE-ACETYLPROMAZINE COCKTAIL-MOUSE

The following regimen will provide a surgical plane of anesthesia for 30-40 minutes and sedation for 1-3 hours.

Mix together: [1.0 ml] Ketamine (100 MG/ML)
[0.5 ml] Xylazine (20 MG/ML) – {BE careful to verify it is 20 mg/ml & not 100MG/ML}
[0.3 ml] Acepromazine (10 MG/ML)
ADD 8.2ml Normal saline 0.9% or PBS (Use Sterile Solution & Store in Sterile Container

Mixtures of ketamine and xylazine are not stable and may lose potency. Make FRESH Mixture WEEKLY. Date of mixing should be placed on container or bottle used.

DOSE: 0.1 ML per 10 grams Body Weight
Delivered Dose is Ketamine 100 mg/kg **/** Xylazine 10mg/kg **/** Acepromazine 3mg/kg
Route:Intraperitoneal injection using 1 ml syringe 23-25 gauge 5/8 inch needle

(3) Tribromoethanol (aka: Avertin, TBE) This anesthetic has been associated with complications from the reconstituted solution. Reconstituted solutions should be used within 2 weeks of thawing because of susceptibility to temperature and light induced changes.
The use of Avertin requires IACUC approval. Avertin is not available as a Pharmaceutical or Medical Grade compound. The use of NON-PHARMACEUTICAL GRADE compounds in animal research is addressed in: IACUC Policy: **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research**. Non-pharmaceutical grade chemical compounds may be used for scientific investigation provided that a scientific justification is provided. Acceptable reasons for use of non-pharmaceutical or chemical grade agents may be;
1. Scientific necessity
2. Non-availability of an acceptable veterinary or human pharmaceutical-grade product.

(4) Alphachloralose provides very little analgesia & should not be used as a surgical anesthetic unless scientifically justified such as may be necessary in certain physiological recording experiments. It may be combined with Urethane and used in non-recovery procedures of long duration where preservation of autonomic reflexes is essential. A Pharmaceutical grade (USP) compound must be used if available (refer to IACUC policy **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research).**

(5) Urethane may be used for non-recovery procedures of exceptionally long duration where preservation of autonomic reflexes is essential. It is often combined with Alphachloralose as an adjunct in long, non-recovery recording procedures. A Pharmaceutical grade(USP) compound must be used if available (refer to IACUC policy **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research).**

(6) Ether may be conditionally acceptable only if approved by the IACUC and EH&S. Scientific Justification must be addressed in the IACUC protocol. EH&S must approve the location, storage and use.

(7) Refer to IACUC Policy No.9906: Euthanasia for guidelines for the humane use of CO_2 for euthanasia of rodents. The IAS euthanasia facilities conform to current AVMA guidelines to displace 20% of the mouse containers air per minute. If using CO_2 in your laboratory, a flow meter must be attached to the system to confirm that the air displacement rate is 20% of the volume of the container per minute.

(8) IACUC approval is required. Adequate scientific justification must be provided for omitting the use of an anesthetic or sedative. Literature searches for alternative methods of humane euthanasia are required as part of the IACUC protocol.

RATS			
	Drug(s) or Method	**Dose (mg/kg)**	**Route**
Anticholinergics	Atropine	0.02-0.05	SC,IM,IV
	Glycopyrrolate	0.5	SC, IM
SEDATIVES / IMMOBILIZATION	Acetylpromazine	2.5	IM, IP
	Diazepam	2.5 - 5	IP,IM
	Xylazine	2 - 8	IM,IP
	Ketamine	50-100	IM,IP
INJECTABLE ANESTHESIA	Ketamine / Xylazine	75-100K / 10X	IP*
	Ketamine-Xylazine mixture [1]	0.15 ml/100gm BW	IP
	Ketamine / Xylazine / Acetylpromazine	40-50 / 2.5 / .75	IM
	Ketamine-Xylazine-Acetylpromazine mix [2]	0.1 ml/100gm BW	IM*
	Ketamine / Diazepam	20-80 / 5-10	IP
	Pentobarbital	30-60	IP, IV
	Avertin (Tribromoethanol)[3]	240	IP
	Alpha-Chloralose[4](Non-Survival)	55-65	IP
	Urethane[5] (Non-Survival)	1000	IP
INHALANT ANESTHETIC	Isoflurane (to effect)	2-5%	inhalant
	Ether[6]	To Effect	inhalant
Physical anesthesia	chilling (neonatal: 1-3 days of age)		ice/water
Analgesia	Carprofen	5	SC
	Flunixin (Banamine) q 12 hr	2.5	**SC**
	Ketoprofen	5	SC
	Meloxicam	1	SC
	Butorphanol (q 4 hr)	1-2	SC
	Buprenorphine (q 12 hr)	0.01 -0.05	SC
	Acetaminophen	200	PO

Euthanasia			
Acceptable	Carbon Dioxide (To Effect)[7]	10-30% Volume Displacement per minute	inhalant
	Inhalant Anesthetic Overdose	to effect	inhalant
	Pentobarbital	>120	IV,IP
	Injectable Anesthetic Overdose	3X anesthetic dose	IV,IP
	Decapitation (Under Anesthesia)		
	Cervical Dislocation (Under Anesthesia)		
Conditionally Acceptable Euthanasia [8]	Exsanguination (Under Anesthesia)		
	Cervical Dislocation (< 200g)		
	Decapitation		

Abbreviations:

IM = intramuscular
IV = intravenous
BW = body weight
SC = subcutaneous
IP = intraperitoneal
q = every

1) KETAMINE-XYLAZINE COCKTAIL -RATS

The following regimen will produce a surgical level of anesthesia for 15-30 minutes and sedation of 1-2 hours.

Combine: [1.0ml] Ketamine (concentration: 100mg/ml)
[0.5ml] Xylazine (concentration: 20mg/ml)

Mixtures of ketamine and xylazine are not stable and may lose potency. Make FRESH Mixture WEEKLY. Date of mixing should be placed on container or bottle used.

DOSE: 0.15ml per 100 gm of Body Weight

Delivered Dose is Ketamine 100 mg/kg **/** Xylazine 10mg/kg
Route:Intraperitoneal injection using 1 ml syringe 23-25 gauge 5/8 inch needle

(2) KETAMINE-XYLAZINE-ACETYLPROMAZINE COCKTAIL-RATS

The following regimen will provide a surgical plane of anesthesia for 30-40 minutes and sedation for 1-3 hours.

Mix together: [0.5 ml] ketamine (100 MG/ML)
[0.125 ml] xylazine (**20** MG/ML) – {BE careful to verify it is 20 mg/ml & not
[0.075 ml] acepromazine (10 MG/ML)
[3.0 ml sterile saline]

Mixtures of ketamine, xylazine & Acepromazine are not stable and may lose potency. Make FRESH Mixture WEEKLY. Date of mixing should be placed on container or bottle used.

DOSE: 0.10 ml per 100 grams Body Weight

Delivered Dose is Ketamine 50 mg/kg **/** Xylazine 2.5mg/kg **/** Acepromazine 0.75mg/kg
Route:Intramuscular injection using 1 ml syringe 23-25 gauge 5/8 inch needle

3) Tribromoethanol (aka: Avertin, TBE) This anesthetic has been associated with complications from the reconstituted solution. Reconstituted solutions should be used within 2 weeks of thawing because of susceptibility to temperature and light induced changes.

The use of Avertin requires IACUC approval. Avertin is not available as a Pharmaceutical or Medical Grade compound. The use of NON-PHARMACEUTICAL GRADE compounds in animal research is addressed in: IACUC Policy: **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research**. Non-pharmaceutical grade chemical compounds may be used for scientific investigation provided that a scientific justification is provided. Acceptable reasons for use of non-pharmaceutical or chemical grade agents may be;

1. Scientific necessity
2. Non-availability of an acceptable veterinary or human pharmaceutical-grade product.

(4) Alphachloralose provides very little analgesia & should not be used as a surgical anesthetic unless scientifically justified such as may be necessary in certain physiological recording experiments. It may be combined with Urethane and used in non-recovery procedures of long duration where preservation of autonomic reflexes is essential. A Pharmaceutical grade (USP) compound must be used if available (refer to IACUC policy **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research).**

(5) Urethane may be used for non-recovery procedures of exceptionally long duration where preservation of autonomic reflexes is essential. It is often combined with Alphachloralose as an adjunct in long, non-recovery recording procedures. A Pharmaceutical grade(USP) compound must be used if available (refer to IACUC policy **2012-01 The Use of Non-Pharmaceutical Grade Compounds in Animal Research).**

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(7) Refer to IACUC Policy No.9906: Euthanasia for guidelines for the humane use of CO_2 for euthanasia of rodents. The IAS euthanasia facilities conform to current AVMA guidelines to displace 20% of the mouse containers air per minute. If using CO_2 in your laboratory, a flow meter must be attached to the system to confirm that the air displacement rate is 20% of the volume of the container per minute.

(8) IACUC approval is required. Adequate scientific justification must be provided for omitting the use of an anesthetic or sedative. Literature searches for alternative methods of humane euthanasia are required as part of the IACUC protocol. Exsanguination under anesthesia must be addressed in the IACUC protocol as “Non-Survival-Surgery”.

RABBITS

	Drug(s) or Method	Dose (mg/kg)	Route
Anticholinergics	Atropine [1]	0.2-2.0	SC,IM,IV
	Glycopyrrolate	0.1	IM
Sedatives	Acetylpromazine	0.5-10	IM,SC
	Ketamine	15-50	IM,SC
	Xylazine	3-6	IV, IM
	Diazepam	5-10	IM
	Diazepam	1-2	IV
Anesthetics	Ketamine / Xylazine [2]	35/ 5	IM
	Ketamine / Xylazine	10/3	IV
	Ketamine / Acetylpromazine	50/1	IM
	Ketamine / Diazepam	25 / 5	IM
	Ketamine / Xylazine / Acetylpromazine	35/5/1	IM
	Ketamine / Xylazine / Butorphanol	35/5/ 0.1	IM
	Pentobarbital	30-45	IV
	Isoflurane (To Effect)	1-5%	inhalant
Analgesics	Buprenorphine q 8-12 Hr	0.01 -.005	SC, IV
	Butorphanol (q 4 Hr)	0.1 - 0.5 I	IV, SC
	Carprofen (Once-Twice Daily	1.5	PO
	Flunixine (Banamine) (q 12 Hr)	1-2	SC,IM
	Ketoprofen	3	IM
	Meloxicam	0.6 - 1	SC

Euthanasia			
Conditionally Acceptable [3]	Pentobarbital	> 100	IV
	inhalant anesthetic OD (to effect)	(to effect)	inhalant
	carbon dioxide (neonates;)	to effect	inhalant
	cervical dislocation (< 1 kg)		
	decapitation (under anesthesia)		
	exsanguination (under anesthesia)		

(1) rabbits produce atropinase, shortening the effects of atropine to 10-20 minutes
(2) may be contraindicated in New Zealand Black rabbits
(3) scientific justification needed; must be considered and approved by Animal Institute Committee; may be acceptable if used in combination with other methods and/or under anesthesia

HAMSTERS			
	Drug(s) or Method	Dose (mg/kg)	Route
Anticholinergic	Atropine	0.04	SC
	Glycopyrollate	0.5	IM
Sedative	Acepromazine	2.5	IP
	Diazepam	5	IM,IP
	Medetomidine	30-100	SC,IP
	Xylazine	1-5	IP
	Ketamine	50-100	IP
Anesthetic	Ketamine (Dilute To 10mg/Ml)	40-100	IM ,IP
	Pentobarbital (Dilute To 6-10 Mg/Ml)	35-90	IP
	Ketamine / Xylazine	200 / 7-10	IP
	Ketamine / Diazepam	70 / 2	IP
	Ketamine / Medetomidine	100 / 0.25	IP
	Ketamine / Acetylpromazine	150 / 5	IP
	Urethane (Non-Survival Only)	1-2 gm	IP
	Isoflurane (To Effect)	1-3%	inhalant
Analgesic	butorphanol (q 4-12 hr)	0.125-2	IM, SC
	buprenorphine (q 8-12 hr)	0.005-2	SC

Euthanasia			
	pentobarbital	> 120	IV,IP
	carbon dioxide (to effect)	10-30% Volume Displacement per minute	inhalant
	inhalant anesthetic OD	(to effect)	inhalant
Conditionally Acceptable [(1)]	decapitation (under anesthesia)		
	exsanguination (under anesthesia)		

(1) scientific justification needed; must be considered and approved by Animal Institute Committee; may be acceptable if used in combination with other methods and/or under anesthesia

PRIMATES -*Macaca fascicularis*			
	Drug(s) or Method	**Dose (mg/kg)**	**Route**
Anticholinergics	Atropine	0.04-0.1	SC,IM,IV
	Glycopyrrolate	0.005-0.01	IM
Sedatives	Ketamine	5-25	IM,IV
	Acetylpromazine	0.2	IM,SC
	Xylazine	0.5	IM
	Diazepam	1	IM, IV
Anesthetics	Ketamine / Xylazine	10 / 0.5	IM
	Ketamine / Diazepam	15 / 1	IM
	Ketanine / Medetomidine	5 / 0.05	IM
	Pentobarbital	20-35	IV
	Isoflurane (To Effect)	1-5%	inhalant
Analgesics	Butorphanol (q 6-8 Hr)	0.01-0.5	IM, SC
	Buprenorphine (q 8-12 Hr)	0.005-1.0	IM, IV,SC
	Flunixin Meglumine (q 8-24 Hr)	41642	IM
	Acetominophen q 8h	6	PO
	Aspirin (q 6-8 Hr)	12.5-20	PO
	Carprofen (Once Daily)	3-4	SC
	Ibuprofen (q12h)	7	PO
	Ketoprofen (Once Daily)	2	SC,IM
	Meloxicam (Once Daily)	0.1 - 0.2	SC,PO

Euthanasia			
Acceptable	Pentobarbital OD	> 100	IV
	Inhalant Anesthetic OD	(to effect)	inhalant
Conditionally Acceptable	Exsanguination (Under Anesthesia) (1)		

(1) must be included in IACUC protocol as Non-Survival Surgery & approved by Animal Institute Committee;

AVIANS			
	Drug(s) or Method	**Dose (mg/kg)**	**Route**
Anesthetic	isoflurane (to effect)	0.25-5%	inhalant
	ketamine /xylazine	20-50 Ketamine / 2-10 Xylazine	IM
	ketamine / diazepam	10-50 Ket / 0.5 -2.0 Diazepam	IM
Analgesic	Acetaminophen (in drinking water)	5mg/L	PO
	buprenorphine (q 2-6hr)	0.05 - 1	SC,IM
	butorphanol (q 1-4 hr)	0.5 - 4	IM, IV
	Carprofen (q 12-24h)	1-10	PO, IM, IV
	Flunixin (q 24h)	1-10	IM, IV
	Meloxicam (q 24h)	0.1 -0.2	PO, IM

Euthanasia

ACCEPTABLE	pentobarbital	> 100	IV,IP
	inhalant anesthetic OD (to effect)		inhalant
	Carbon Dioxide	20% volume displacement per minute	inhalant
Conditionally Acceptable [(1)]	cervical dislocation		
	decapitation (under anesthesia)		
	exsanguination (under anesthesia)		

(1) must be included in IACUC protocol as Non-Survival Surgery & approved by Animal Institute Committee

AMPHIBIANS

	DRUG(S) OR METHOD	DOSE (MG/KG)	Route
Sedative	ketamine	50-150	SC,IM
Anesthetic	tricaine methanesulfonate * (larvae, newts)	200-500 mg/L	immersion
	" (frogs, salamanders)	500mg-2 g/L	immersion
	" (toads)	up to 3 g/L	immersion
	" (sterile) (frogs)	100-400	immersion
	benzocaine *	50 mg/L	immersion
	" (frogs, salamanders)	200-300 mg/L	immersion
	isoflurane	4-5%	inhalant
Analgesic	buprenorphine	99.1 nM/g	SC
	meperidine	128.1 nM/g	SC
Euthanasia			
(Acceptable)	carbon dioxide (to effect)	100%	inhalant
	inhalant anesthetic OD (to effect)		inhalant
	tricaine methanesulfonate OD (to effect) (1)*		immersion
	benzocaine OD (to effect)*		immersion
Conditionally Acceptable (1)	double pithing		
	decapitation (under anesthesia, followed by pithing)		

* Solutions of Benzocaine & Tricaine Methane Sulfonate (MS-222) must be buffered when used in high concentrations according to the most recent AVMA guidelines for humane euthanasia.

(1) must be included in IACUC protocol as Non-Survival Surgery & approved by Animal Institute Committee

FISH			
	Drug(s) or Method	**Dose (mg/kg)**	**Route**
Sedative	tricaine methanesulfonate (MS-222)*	50 mg/L	immersion
Anesthetic	benzocaine*	25-50 mg/L	immersion
	tricaine methanesulfonate (Fresh Water ONLY)	100 mg/L	immersion
	ketamine	14-18	IM
	etomidate	2-4 mg/L	immersion
	metomidate	2.5-5 mg/L	immersion
Euthanasia			
Euthanasia	pentobarbital	>100	IV,IP
	tricaine methanesulfonate OD (to effect)*	1-3 g/L	immersion
	benzocaine * OD (to effect)	100 mg/L	immersion
Conditionally Acceptable(1)	decapitation (under anesthesia, followed by pithing))		

* Solutions of Benzocaine & Tricaine Methane Sulfonate (MS-222) must be buffered when used in high concentrations according to the most recent AVMA guidelines for humane euthanasia.

(1) must be included in IACUC protocol as Non-Survival Surgery & approved by Animal Institute Committee

REFERENCES

Major References:

E.Fish, M.J.Brown, and P.J.Danneman, A.Z Karas,Eds. (2008). Anesthesia and Analgesia in Laboratory Animals (Second Edition. American College of Laboratory Animal Medicine Series, Academic Press, San Diego.

Flecknell, PA (2009). Laboratory Animal Anesthesia (Third Edition). Academic Press, Harcourt Brace Jovanovich, London.

Hawk, CT and SL Leary (1995). Formulary for Laboratory Animals. Iowa State University Press, Ames.

AVMA Panel on Euthanasia (2013). 2013 report of the AVMA panel on euthanasia. Journal of the American Veterinary Medical Association.

Additional References:

Close, B, K Banister, V Baumans, et al. (1997) Recommendations for euthanasia of experimental animals: Part 2. Laboratory Animals 31:1-32.

Spikes, SE, SL Hoogstraten-Miller, GF Miller (1996). Comparison of five anesthetic agents administered intraperitoneally in the laboratory rat. Contemporary Topics 35:53-56.

Hubbell, JAE, WW Muir (1996). Evaluation of a survey of the diplomates of the American College of Laboratory Animal Medicine on use of analgesic agents in animals used for biomedical research. Journal of the American Veterinary Medical Association 209(5):918-921.

Hansen, BD (1994). Analgesic therapy. The Compendium (Small Animal) 868-873.

Rush, HG (1996). Interpretive study of the report of the AVMA panel on euthanasia. Contemporary Topics 35(2):42-44.

Danneman, PJ and TD Mandrell (1996). Evaluation of six anesthetic regimens in neonatal rats. Contemporary Topics 35(4):60.