

INSTITUTIONAL ANIMAL CARE AND USE COMMITTEE (IACUC)

GUIDELINES FOR PREPARING DRUGS AND AGENTS FOR ANIMAL RESEARCH

PURPOSE

The purpose of this document is to provide guidelines for preparing drugs and research agents for administration to animals.

DEFINITIONS

Aliquots: material transferred to secondary containers, and not diluted or mixed.

Diluent/solvent/vehicle: carrier or inert medium used as a solvent (or diluent) in which the medicinally active agent is formulated and or administered.

Dilution: stock material mixed with diluent/vehicle to produce required concentration.

Food grade: material certified as safe for human or animal consumption.

Formulation: different chemical substances, including active drug, combined to produce a final product.

Reconstituted solution: medication supplied in powder form mixed with liquid before administration.

Single-use vials: a vial of liquid medication intended for parenteral administration (injection or infusion) that is meant for use in a single patient for a single case, procedure, injection. Single-dose or single-use vials are labeled as such by the manufacturer and typically lack an antimicrobial preservative.

USP grade: a chemical grade of sufficient purity to meet or exceed requirements of the U.S. Pharmacopeia (USP); acceptable for use in the manufacture of food, drugs, or medications; may be used for most laboratory purposes.

GUIDELINES

1. Commercially available human or veterinary substances or formulations should be used whenever available. Agents for sedation, analgesia, anesthesia, or euthanasia should be commercially available veterinary or human preparations, unless the use of an investigational chemical or formulation is scientifically necessary, appropriately justified, and approved by the IACUC.

2. When formulating from bulk drug or chemical, US Pharmacopeia (USP) grade material should be used when available. If USP grade drug cannot be obtained, material of the of the highest purity available, e.g., American Chemical Society (ACS) grade or reagent grade, should be used.
3. Diluents, vehicles, and/or solvents must be specified in the animal use protocol. Biocompatible diluents, solvents, and vehicles, preferably of USP grade and sterile, should be used. The vehicle(s) used can limit drug concentration, dose volume, and route of administration; for information on specific vehicles, see Gad, et. al. (2006). Information on appropriate volumes of administration can be found on the [Veterinary Guidance](#) section of the CCM website, under “Substance Administration”.
4. When formulating compounds for oral use, food-grade materials should be used.
5. When formulated for parenteral administration (e.g., IV, IP, IM, SC, etc.), compounds must be prepared in an appropriate manner.
 - Sterile constituents (e.g., bulk powder, diluents, solvents, and/or vehicles) must be used, unless not available (e.g., new investigational substances).
 - Compounds must be prepared using aseptic technique. Preparation in a HEPA filtered laminar flow hood (biosafety cabinet, tissue culture hood, animal transfer station) is recommended.
 - The final product must undergo sterilization prior to injection. Sterilization can be accomplished by passage through a 0.22 μm filter, by autoclaving, or by gamma irradiation. Justification must be provided if a larger filter (e.g., 0.45 μm) must be used or if the product cannot be sterilized.
 - The final product must be stored in a sterile vial or tube with a rubber seal to maintain sterility. A sterile vial or tube with a screw cap is acceptable if the product will be used only on the day it is prepared. See Appendix 1 for examples.
6. pH of solutions for parenteral injection should be between pH 4.5 and 8.0. Use of a solution with a pH outside this range should be addressed in the animal use protocol.
7. The osmolality of intravenously administered compounds should generally be formulated to a range matching the research species’ blood osmolality. Generally, agents with an osmolality greater than 600 mOsm/kg cause red blood cell crenation while agents less than 150 mOsm/kg will cause hemolysis, both of which are associated with pain and physiologic disturbances. Solutions with an osmolality less than 450 mOsm/kg are generally well tolerated by intravenous infusion in most research mammals.
8. Containers must be labeled as described in [Labeling Requirements and Expiration Dates for Research Drugs](#). Label templates can be found on the [IACUC Policies & Guidance](#) section of the IACUC website, under “Guidance and Standards”.
 - Contents
 - Concentration

- Date of reconstitution/preparation
- Expiration date
- Initials of preparer

Example 1: [Controlled Substances Label](#)

KX2		
1 mL = Ketamine (50 mg), Xylazine (5 mg)		
Species:	Dose (mL/kg):	
Prep:	Exp:	Made by:

Example 2: [Other Substances Label](#)

Heparinized Saline	
20 units Heparin + 0.9% NaCl/mL	
Prep Date:	
Exp. Date:	
Made by:	

9. Recommended shelf life/expiration dates

- **Unopened drugs or agents in the original packaging:** expiration/use by date printed on the label or packaging
- **Opened single-use/single dose vials:** a single-dose or single-use vial that has been opened or accessed (e.g., needle-punctured) the vial should be discarded according to the time the manufacturer specifies for the opened vial or at the end of the case/procedure for which it is being used, whichever comes first. It should not be stored for future use.
- **Opened multi-dose vials/bags of injectables:** a multi-dose vial/bag that has been opened or accessed (e.g., needle-punctured) should be dated and discarded within 28 days unless the manufacturer specifies a different (shorter or longer) date for that opened vial/bag.
- **Aliquots:** expiration date/use by date printed on the label or packaging of stock material.
- **Reconstituted solutions:** expiration date/use by date/instructions printed on the label or packaging. Reconstituted solutions must be stored under the conditions described in the packaging.
- **Dilutions:** discard 28 days after preparation
- **Mixtures / formulations:**
 - 28 days after preparation, sooner if the mixture becomes clouded or precipitates. Any mixture that becomes clouded or precipitates should not be used in animals and should be discarded immediately.
 - If the stability of an agent in formulation is unknown, it should be prepared fresh prior to each use.

10. Agents should be stored appropriately to retard contamination and degradation (e.g., freezer, refrigerator, etc.). Controlled substances, including dilutions and mixtures containing controlled substances, must be stored in a securely locked, substantially constructed double-locked cabinet or safe in locations where access is limited, except when in use by authorized personnel. Controlled substances that must be refrigerated should be stored in a locked box within a locked refrigerator and the refrigerator should be behind a locked door. (see [MGH Policy on Controlled Substance Use in Non-Human Research](#)).
11. Expired agents must be disposed of properly. If not discarded immediately, expired containers must be labeled “expired” and stored separately from unexpired agents.

Please note: Pyrogens, such as endotoxins, may cause fever or physiological shock when injected into an animal. Pharmaceutical agents are tested for pyrogens. Sterility does not assure that pyrogens are not present. Filtering does not remove pyrogens. Pyrogenicity is a potential experimental variable that researchers should be aware of when using non-pharmaceutical grade agents.

RELATED POLICIES

[IACUC Policy on the Use of Non Pharmaceutical Grade Substances](#)
[MGH Policy on Controlled Substance Use in Non-Human Research](#)

REFERENCES

AAALACi. [Frequently Asked Questions, C. Institutional Responsibilities, 9. Non-Pharmaceutical-Grade Compounds](#). June 2017.

Gad, S.C.; Cassidy, C.D.; Aubert, N.; Spainhour, B.; and Robbe, H. [Nonclinical Vehicle Use in Studies by Multiple Routes in Multiple Species](#) *International Journal of Toxicology* 25:499-521, 2006.

U.S. Department of Health and Human Services. [Drug Packaging and Labeling Control - Expiration Dating](#). Code of Federal Regulations (CFR), Title 21, Volume 4, Chapter 1, Subchapter C, Part 211.137. 2020.

U.S. Department of Health and Human Services. [FAQs Regarding Safe Practices for Medical Injections - Questions about Multi-dose Vials](#) Centers for Disease Control and Prevention: June 20, 2019.

U.S. Department of Health and Human Services. [FAQs Regarding Safe Practices for Medical Injections - Questions about Single-dose/Single-use Vials](#) Centers for Disease Control and Prevention: June 20, 2019.

U.S. Department of Health and Human Services. [Guidelines for the Use of Preservative-Free Pharmaceuticals and Parenteral Fluids in Laboratory Animals](#) National Institutes of Health, Animal Research Advisory Committee: Bethesda, MD, April 27, 2016.

U.S. Food and Drug Administration. [Approved Animal Drug Products - Green Book](#)

U.S. Food and Drug Administration. [Orange Book: Approved Drug Products](#)

v1.1, 21 April 2021

Appendix 1. Recommended storage containers for aliquots, dilutions, or mixtures/formulations of drugs or fluids

For drugs or agents that will be prepared and used on the same day, sterile screw cap vials or tubes like the ones pictured below may be used.



If drugs or agents will be used for more than one day, a sterile container with a rubber septum should be used. An additive free Vacutainer is an acceptable alternative.

