Purpose:

This SOP outlines how to prepare injectable anesthetic ketamine and xylazine cocktails which can be used for surgical plane of anesthesia in rats.

Materials

- Ketamine 100 mg/ml
- Xylazine 100 mg/ml
- Atipamezole 5 mg/ml
- Sterile saline or sterile water
- 10 ml sterile glass empty vial

Mixing Instructions for Delivery of 91mg/kg Ketamine/9.1mg/kg Xylazine:

- Verify the concentration of your drugs prior to mixing
- For an 11ml final volume using ketamine 100mg/ml and xylazine 100 mg/ml add:
  - 10 ml ketamine (100 mg/ml)
  - 1.0 ml xylazine (100 mg/ml)
  - Into a sterile 10ml empty vial
- Use the following template for label:
  - Rat Anesthetic Mix: Ketamine/Xylazine
  - Dosage: 0.1 ml/100 gm IP
  - Delivers **91 mg/kg Ketamine/9.1 mg/kg Xylazine**
  - Concentration: 91 mg/ml Ketamine/9.1 mg/ml Xylazine
  - Expires: 90 days from mixing date

Expiration Date:

- The expiration date for the cocktail is determined by either 90 days from date of mixture or the manufacturer date on either of the drugs being mixed together, whichever comes first.
- Mixtures of ketamine and xylazine can lose potency after mixing, if any indication of this (precipitation, color change), discard immediately and make fresh.

<u>Instruction for use if animal needs a "bump" of anesthesia:</u>

- Re-dosing of this combination of drugs for long procedures >1-1.5 hour is not recommended
- Use of isoflurane gas is strongly recommended for maintenance after injectable drugs (>30-45 min)
- If re-dosing is necessary, **use ketamine only** at 1/3-1/2 half of the original dose (do not exceed 50mg/kg). Make a 1:10 dilution of ketamine only in sterile water or saline (10 mg/ml).

<u>Reversal of Xylazine with Atipamazole:</u>

Both xylazine and dexmedetomidine are potent sedatives and can lead to long recover times if not reversed. Both drugs are reversible with the injectable product atipamazole (e.g. Antisedan® 5 mg/ml). However, ketamine is not reversible following injection, but must be metabolized thus we recommend using reversal at least after 10-20 minutes after anesthesia.

Inject 1 mg/kg either subcutaneously or IP for reversal of the sedative and analgesic effects.

1. Dilution Instructions for delivery of 1 mg/kg Atipamazole
    - Verify the concentration of your drug prior to mixing
    - Dilute atipamazole to 1 mg/ml:
        - For a 2.5 ml vial using atipamazole 5 mg/ml:
            - 0.5 ml Atipamazole (5 mg/ml)
            - 2.0 ml Sterile Saline
            - Using this dilution, dose rats at 0.1 ml/100g body weight SQ or IP. Dose is administered as 1 mg/kg
            - Expiration date: 90 days from mixing date

Rat Weight	**Dosage 1 mg/kg**
100 g	0.10 ml
125 g	0.12 ml
150 g	0.15 ml
175 g	0.18 ml
200 g	0.20 ml
225 g	0.22 ml
250 g	0.25 ml
275 g	0.28 ml
300 g	0.30 ml
325 g	0.32 ml
350 g	0.35 ml
375 g	0.38 ml
400 g	0.40 ml
450 g	0.45 ml
500 g	0.50 ml

Reference

https://www.ncbi.nlm.nih.gov/pmc/articles/PMC2786925/ "Beyond-Use Dating of Extemporaneously Compounded Ketamine, Acepromazine, and Xylazine: Safety, Stability, and Efficacy over Time" J Am Assoc Lab Animal Sci 2009 Nov; 48 (6)

Revision History:

Revision	**Author**	**Revisions Made**	**Replaces version**
1	GC	Formatting and addition of Figure 1	10/29/18
2	GC JD	Removal of multiple dosages, add table-reformat; CVS Review	2/25/20
3	GC	Add SQ route and changes dosage and duration to match Vet Guidelines	10/27/22
4	GC/JD	Addition of Atipamazole dosing, removal of Fig 1 and duration of anesthesia of K/X	5/28/23