

# **Mouse Retro Orbital Injection**

It is recommended that mice be an esthetized for retro-orbital (RO) injection. Mice do not need to be in a surgical plane of an esthesia but need to be sedated deeply enough to be completely immobilized. This aids in proper performance of the technique and minimizes the possibility of ocular injury.

## **Necessary Supplies**

- 27 to 30 gauge needles and syringes
- Isoflurane anesthetic equipment
- Optional topical anesthetic (0.5% Proparacaine hydrochloride ophthalmic solution)

### **Technique**

- 1. Samples will be administered into the venous sinus located behind the orbit. Review the anatomy to be familiar with the area (Image 1). Anesthetize the animal in the induction chamber until it is fully immobilized.
- 2. Lay the animal on its side. Maintenance of anesthesia on a nose cone with 1-3% isoflurane is typically needed until your level of proficiency allows you to perform the procedure before the animal wakes up.
- 3. Retract the eyelids until the globe protrudes slightly out of the orbit and creates a space behind the globe to access the sinus. This will ensure that you do not insert the needle into the globe. (Image 2)
- 4. Make sure there are no air bubbles in the syringe prior to injection.
- 5. Insert the needle at the medial/nasal corner of the eye using a 30-45° angle. Keep the bevel facing down to avoid injuring the globe. (Image 3)
- 6. Insert the needle 2 3mm until you feel contact with bone. Slightly withdraw the needle away from the bone and slowly inject the compound. After injection, slowly withdraw the needle. There should be little to no bleeding.
- 7. If general anesthesia is used, animals must be appropriately monitored until recovered from anesthesia.

### **Notes:**

- Use a new needle per animal
- Maximum recommended volume to be injected is 200 uL (or 0.2mL) per adult mouse (>25g body weight).
- This is an easier alternative for intravenous (IV) tail injection and may be a suitable alternative given your specific scientific aims.



**Image 1-** Anatomical position of the retro orbital venous plexus.



**Image 2-** Eyelid retraction to induce slight proptosis of the globe.



**Image 3-** IV administration into the retro orbital sinus.

### Additional References: