Lateral Tail Vein Injection in Mice and Rats (Preferred Technique For Vascular Access in Mice) IACUC Standard Procedure Effective Date: June 2016

Description of procedure:
The most accessible vessels for intravenous administration in rodents are veins that run the length of both lateral aspects of the tail. Only properly trained personnel can perform this procedure.

Supplies:
- Sterile 27-30 gauge needles for mice
- Sterile 25-27 gauge needles for rats
- 300-500 microliter syringes for mice
- 1ml or 3ml syringes for rats
- Heating device
- Gauze sponges
- Isoflurane Anesthesia System (recommended)

Note: It is highly recommended that new users of this technique anesthetize the animals while conducting this procedure until proficiency is obtained.

Procedure Steps:
1. Weigh each animal before injection. Up to 1% of the animal's body weight in volume can be administered per injection.
2. Record body weights and agent volumes to be administered for each animal.
3. Prior to injection, warm animal for 5-10 minutes to dilate the veins. Animal may be warmed by placing the animal in a commercially available warming box or by using a warm water circulating pad placed under the cage. These are the safest and most effective ways to warm rodents. If an overhead heat lamp is used, extra care must be taken to prevent overheating the animal.
4. It is recommended to lightly anesthetize the animal.
5. Position the animal on its side. Animal should be placed on a rechargeable heat pack or circulating warm water pad to keep the animal warm during anesthesia.
6. Conscious animals need to be restrained using a commercially available restraint of appropriate size (see below). The duration of the restraint should be kept to a minimum, and the equipment washed frequently to prevent pheromone-induced stress or cross contamination.

**Note:** With brown or black mice and rats, an additional light source may be necessary to aid in visualizing the tail veins. Rats have scales making the vein difficult to see, especially in older adults. The scales are removed by gently cleaning the tail with a saline or chlorhexidine solution making the veins more apparent—wipe in the direction of the scales to avoid irritation to the tail.

**Mouse:** Grasp the tail at mid-length or at the distal (further down the tail) end. The index and middle fingers of the non-dominant hand are placed around the tail above where the needle will be inserted (digital pressure will act as a tourniquet). The lower part of the tail is held between the thumb and ring finger below the injection site. Put slight tension on the tail by applying pressure with both sets of fingers. Release pressure to the proximal fingers before administering the agent into the vein.

**Note:** With mice, elevating the animal about 4-6 inches off the table may be helpful with keeping the needle and syringe parallel to the vein.
**Rats:** A tourniquet is used to constrict the vein. A tourniquet is made with a rubber band wrapped around the top of the tail and held together firmly with a hemostat. The tourniquet is released before the agent is administered into the vein.

7. Hold the syringe with the dominant hand near the bottom so that the remaining fingers are near the plunger and can easily push the agent into the vessel without disrupting the needle in the vein.
8. Syringes should be prepped with no air bubbles. Insert the needle (small gauge, 27-30 for mice and 25-27 for rats), bevel up, into the vein towards the direction of the head. Keep the needle and syringe parallel to the tail. Aspiration is not advised as it may cause the vein to collapse, but a flash of blood in the hub of the needle may be seen when first placed. Proper placement may not be verifiable until injection occurs, but when placed correctly the needle should advance smoothly into the vein.
9. Slowly inject. If there is resistance and/or a blister or white area appears above the needle on the tail, the needle should be removed and re-inserted above the first site.
10. Remove the needle and apply gentle compression until bleeding has stopped.
11. If the animal was anesthetized, monitor the animal during the recovery process.
12. Return animals to their cage and observe to make sure that bleeding has not resumed.

**Agents:**

This procedure recommends anesthesia. All agents administered to animals should be listed in the "Agents" section of the RIO IACUC protocol.

**Adverse Effects:**
Adverse effects should be listed in the “Adverse Effects” section of the RIO IACUC protocol.

Examples of potential adverse effects include: Peri-vascular irritation, Blood loss