Title: Intravenous Tail Vein Injections  
SOP Number: 067  
Purpose: This Standard Operating Procedure describes the methods for the intravenous injection in rodents using the lateral tail veins.

Equipment Required:
1. Sterile needle and syringe
   A. 27-30 gauge needle for mice
   B. 25-27 gauge needle for rats
2. Mouse restraining device
3. Heating device
4. Alcohol pads
5. Gauze
6. Anesthesia machine

Note: Anesthesia is not required for this procedure but is recommended to users that are unable to perform this technique on conscious animals.

Procedure:
1. 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 heating pad placed under the cage. These are the safest and most effective ways to warm rodents. Direct heat can also be placed on the tail using a hand warmer momentarily. If an overhead heat lamp or hand warmer is used, extra care must be taken to prevent overheating the animal or burning the tail.
2. It is recommended to lightly anesthetize the animal with Isoflurane.
3. When the animal is anesthetized, position the animal in lateral recumbency on a safe heat source or heat pad to maintain core body temperature during anesthesia.
4. Conscious animals need to be restrained using a commercially available restraint device of appropriate size (see below). Placing a towel over the restrainer keeps the animal in a dark, quiet place and helps calm the animal. The duration of the restraint should be kept to a minimum, and the equipment washed frequently to prevent pheromone-induced stress or cross contamination. Rodents sometimes spin in the restrainer; be sure to confirm to orientation and location of the lateral tail vein before performing injections.
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 not contain air bubbles. Do not reuse syringes or needles. Needles should be sharp and replaced after three attempts. 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. There is no need to aspirate 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. The needle should advance smoothly into the vein if placed correctly.
9. Inject slowly. If there is resistance and/or a blister or white area appears above the needle on the tail, the needle should be removed. A max volume of 0.2ml in mice and 0.5ml in rats is acceptable.

Note: Starting low on the tail allows for operator error as the user can move above the first injection site for additional attempts.

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.

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. Needle should enter the vein at a shallow depth, keeping syringe and needle parallel to tail. Release pressure to the proximal fingers before administering the agent into the vein. No resistance should be felt when depressing the plunger. 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 to allow visualization and access to the vein for injection mid-length or at the distal (further down the tail) end. 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.

Examples of potential complications include: Extravasation and peri-vascular irritation or tail necrosis, Blood loss

References:
Hooke Laboratories; injection angle image.


Animal Health Monitoring:
1. Animals are observed daily by animal care staff for any evidence of illness or change in behavior.
   A. Everyone with access to the animal facility is responsible for immediately informing the facility manager or a university veterinarian when an animal becomes ill or a change in behavior is observed.
2. In the event of suspected illness:
A. Record your observation on the treatment/observation sheet- include the date, animal #/cage ID, the problem observed, and initials

B. Contact the ACS facility manager or a university veterinarian:

   Dr. Tiffanie Brooks, Attending Veterinarian
   806-834-8588 Office
   806-239-2120 Cell Phone

   Paul Stonum, Clinical Veterinarian
   806-834-7373 Office
   660-562-4425 Cell Phone

   Sydnee Woodman, Manager
   806-834-2872 Office
   602-758-0670 Cell Phone