Purpose:

- The purpose of this workshop is to inform and instruct personnel on the safe and the least stressful way to restrain and inject mice and rats. This workshop will illustrate proper restraint while administering injections to commonly used sites.
Proper handling and restraint is one of the most important factors in being able to give an injection properly while limiting the amount of stress and/or discomfort to the animal or the handler.
Types of restraint:

- When working with lab animals the two types of restraint that are utilized most often are physical and chemical.
Chemical restraint is recommended if there will be great distress or discomfort to the animal.

This can be achieved by using a gas anesthesia such as isoflurane or by a type of injectable sedation such as Ketamine/Xylazine mix.
Physical restraint can be achieved by many means. Holding the animal with gloved hands is very common. There is also the use of commercial type restrainers that can be purchased from laboratory equipment suppliers. Towels can be used, the wire bar lid can be utilized as well.
Manual restraint of a mouse using both hands. (a) The mouse is placed on the cage lid with the preferred hand. The tail is pulled gently back by the hand. (b) The mouse is quickly and firmly picked up by the scruff of the neck behind the ears with thumb and index finger of other hand. (c) The tail is transferred from the preferred hand to between palm and little or ring finger of the other hand, then held firmly. (d) The mouse is restrained.
Double Handed Restraint
Single-handed restraint of the mouse. (a) The tail is picked up using thumb and forefinger of the preferred hand. (b) The mouse is placed on the cage lid or other solid surface pulling gently back by the hand. (c) The tail is immediately grasped by the palm and middle finger, ring finger and/or little finger and then, the tail held between thumb and forefinger is released. (d) and (e) The fold of skin from the scruff of the neck down the back is immediately gripped using the thumb and forefinger. (f) The mouse is restrained.
Single Handed Restraint
The injection sites that I will be discussing can be used on both mice and rats. These will include Sub-Cutaneous (SQ), Intraperitoneal (IP), and Tail vein injections. Intramuscular (IM) injections are not recommended because of how small the muscles are. I will also briefly touch on needle and syringe sizes, volumes that can be administered, and commonly used restraint.
SubCutaneous

- In both the mouse and the rat SQ injections are usually given in the scruff (access skin) of the neck or using the scruff back by the hind quarters by tenting the skin and making your injection.
SQ Needles, Syringes, and Volumes

- Depending on what is being injected (the viscosity) can help determine the size needle that will be needed. The smallest gauge that can be used is preferable for the comfort of the animal. For a thin substance a small gauge needle (27g, 26g, 25g) can be used. When injecting something like tumor cells you may want to use a larger gauge needle (25g) as to not lyse the cells that are being injected. For SQ injections a max volume of 2–3cc. (Flecknell, 1987, Reeves et al., 1991; Wolfensohn and Lloyd, 1994)
Intraperitoneal

- When giving an IP injection good restraint and good injection technique will help minimize any secondary problems that may occur with this type of injection.
- Restrain your animal using either the scruff and holding the tail with pinky or ring finger in mice. If using rats gently grabbing them over the shoulders causing the legs to cross over the chest to help prevent getting bit is common restraint.
Once animal is restrained turn over so abdomen is exposed. Please monitor chest movements to make sure the animal is doing ok. On the mouse you want to make your IP injection in the lower right or left quadrant of abdomen trying to avoid hitting bladder, liver, or other internal organs.

Supplies:
- Tuberculin 1cc syringe
- 25g needle(s)
- Max volume is 20ml/kg
(Ref. IQ 3Rs Leadership Group– Contract Research Organization Working Group)
When performing the IP injection on the rat you should inject into the lower right quadrant of the abdomen to avoid hitting such organs as liver, bladder, and cecum.

**Supplies:**
1–3cc syringe
21–25g needle(s)
Max amount 10ml/kg
(Ref. IQ 3Rs Leadership Group– Contract Research Organization Working Group)
Intravenous (using lateral tail vein)

- When doing an IV injection using the tail vein it is very convenient to have one of the commercial restraint devices available. This could be the plastic restrainers or DecapiCones.
Once the animal is restrained the tail vein is located laterally on both the right and left side of the tail. You always want to start your injection at the lower portion of the tail ~ 1/3 from the tip. This allows you to move up the tail if the injection was unsuccessful. The tail should be warmed to help dilate the veins. This can be done by using a small heating source or placing tail in warm water for 1–3 minutes.
You have your animal restrained and vessels dilated. The next step take your syringe with needle and substance to be injected. Hold needle parallel to the tail with bevel side up.

Gently insert needle into vein while pulling back on plunger. You will get a flash of blood into needle hub when in the vein. Begin injection if bubble or bleb appears under skin remove needle and inject closer to the base of the tail. Needle needs to be changed if this is done. When needle is removed apply light pressure to stop bleeding.
IV Injections

- Supplies for Mouse:
  - 1cc Syringe
  - 27–30g needle(s)
  - Restraint Tube
  - Heating Source
  - Max Volume 0.2ml
IV Injections

- Supplies for rats:
- 1–3ml syringe
- 25g Needle
- Heat Source
- Max Volume 0.5ml
This concludes the PowerPoint. If you have further questions please feel free to contact the personnel below to help you.

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