Arizona State University Institutional Animal Care and Use Committee STANDARD INSTITUTIONAL GUIDELINE

TERMINAL BLOOD COLLECTION

Terminal blood collection techniques in mice and rats are used when a single, large amount of blood is necessary for experimentation. The techniques can only be performed while the animal is under deep anesthesia or following humane euthanasia. The recommended technique is cardiac puncture, and to use this approach IACUC protocols need only to reference this SIG rather than describe the details of the procedure. Alternative terminal bleed approached include the abdominal aorta or vena cava. Alternative sites such as these may be used if the procedure is thoroughly described in the approved IACUC protocol.

Cardiac Puncture:

- 1. Have all supplies ready before starting, including placing needles on syringes and prepping collection tubes as necessary.
 - a. If plasma is needed, then add heparin (10-30 USP units/mL of blood) to the blood collection tubes. Also, prepare the syringes by drawing up some heparin from a prepped collection tube into the syringe to coat the inside of the syringe, then inject the heparin back into the blood collection tube.
 - b. Use a 20-23 g needle for both mice and rats, but attach it to a 1 ml syringe for mice and a 3 ml syringe for rats.
- 2. The animal is either humanely euthanized using CO₂ or deeply anesthetized as described in the IACUC protocol.
 - a. If using CO₂, remove the animal from the induction chamber as soon as it stops breathing. The animal will stop breathing before the heart fully stops beating, and this will help to ensure the best quality and maximum volume of the sample.
 - b. If using anesthesia, then anesthetic agent and dose to be used needs to ensure the animal maintains appropriate anesthetic depth (i.e., unresponsive to firm toe pinch) during the procedure. Inhalant anesthesia may be used if the animal is maintained under anesthesia using a nose cone during the entire procedure.
- 3. Place animal in dorsal recumbency.
- 4. Insert needle slightly below and to the left of the xyphoid process at a 30-40° angle to the body, aimed towards the animal's head.
- 5. Retract the plunger slightly to create a vacuum inside the syringe and either advance or retract the needle (depending on the depth of the needle) until blood appears in the needle hub.
- 6. While keeping the needle stable, gently retract the plunger until a sufficient amount of blood has been collected.
 - a. If the blood stops flowing before the desired amount is obtained, try gently rotating the syringe as the needle may be abutted against the ventricle wall.
 - b. If the needle moves during the blood collection, it may need to be re-inserted into the heart to continue blood collection.
- 7. If the animal was anesthetized for the procedure, then after the completion of the procedure it needs to be euthanized by bilateral thoracotomy or cervical dislocation. Ensure the animal is still deeply under anesthesia before performing this step.

Parasuraman, S., R. Raveendran, and R. Kesavan. "Blood sample collection in small laboratory animals." *Journal of pharmacology & pharmacotherapeutics* 1.2 (2010): 87.

Joslin, Janis Ott. "Blood collection techniques in exotic small mammals." *Journal of Exotic Pet Medicine* 18.2 (2009): 117-139.