Blood Collection

Site Preparation: Certain general procedures and precautions are applicable to methods of blood collection as well as to administration of fluids and anesthetics. When venipuncture is required, hair should be shaved from the site for better visibility. The area of injection should be cleansed with alcohol. Some procedures will require anesthesia; others may be carried out without anesthesia, provided suitable restraint is possible. In order to visualize veins better, one of several methods of dilation may be used. The vessel may be occluded with digital pressure to cause enlargement. Heat will also cause dilation. When using the rabbit ear, or mouse or rat tail, a low watt light bulb may be used for heat. The preferred method of collection of large volumes of blood from the rabbit ear is with the use of a droperidol-fentanyl tranquilizer that promotes arterial dilation and makes blood collection from rabbits simple for even the inexperienced phlebotomist (Drug Dosage Table -Table 1).

Guidelines for blood collection **or** A Good Practice Guide to the Administration of Substance and Removal of Blood, Including Routes and Volumes
http://www.aaalac.org/accreditation/RefResources/BloodRemoval.pdf

Equipment needed: Needles of appropriate gauge and length must be selected with care. For the tail vein or artery of rats and mice, small needles (25-30g) should be used. For other vessels in other animals, the suitable size will depend upon the size of the animal and vessel.

Technique: Proper insertion of the needle into the vein or artery is the most tedious part of the procedure. Certain guidelines may be given, but only practice can be expected to provide any proficiency. A precise, careful introduction of the needle is always best. The needle is inserted parallel above the vessel and the tip directed into the lumen along with the longitudinal axis. The intracardiac puncture generally represents the most practical method of blood collection from small rodents when more than a few drops are required. It is also useful in rabbits for exsanguination. Animals must be anesthetized and placed in dorsal recumbency. The point of the strongest heart beat is determined with the forefinger. The needle is inserted through the skin, between the ribs at this site, directly into the heart. Blood should be withdrawn slowly. The cardiac route for blood collection is a terminal procedure.

In the rabbit, the marginal ear vein is most useful for intravenous injection, but not blood collection. Blood collection is best accomplished from the central ear artery via butterfly catheter or needle (Figures 10 a & b). Thirty to forty ml may be collected in this manner. The absolute maximum of blood to be withdrawn at one time is 9 ml/kg body weight. The PCV (packed cell volume) must be measured at each collection if such large volumes of blood must be withdrawn. If the PCV drops below 35%, collection must be reduced. The use of a droperidol-fentanyl tranquilizer promotes arterial dilation, relaxes the rabbit and makes blood collection from rabbits simple for even the inexperienced phlebotomist (Table 1). DLAM is available to demonstrate or perform this service.

Figure 10: Central Ear Artery of the Rabbit

In the rodent, blood collection by cutting off toes is not permitted. Collection from the tail artery may be increased by warming it in water. Animals should be restrained in restraining device or anesthetized. After cleaning, a small nick is made on the ventral midline of the tail and blood is collected. Digital pressure will stop the blood flow. Withdrawal of blood from the orbital venous plexus of rats and orbital sinus of mice and hamsters is frequently used. When bleeding the mouse, hamster and rat by the retrobulbar technique, the hematocrit capillary tube is directed toward the major venous structures of the orbit. Knowledge of the location of the venous structures and the technique is essential (Figures 10 &

11). Anesthesia is required for all retrobulbar bleeding procedures. Instruction on all of these blood-collection techniques is available through DLAM.

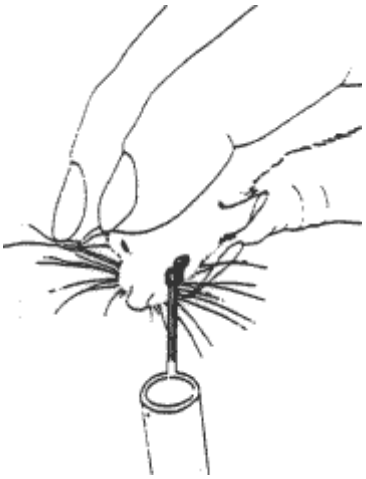
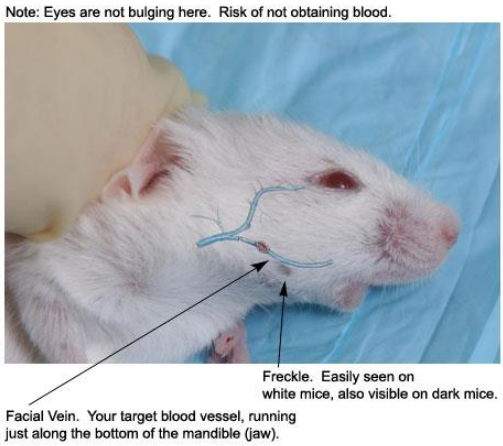


Figure 11: Retrobulbar Blood Sample Collection in the Mouse



Submandibular Blood Collection in Mice

- ✓ Locate the hairless freckle on the side of the jaw.
- ✓ Pick up the sharp instrument of your choice (lancet or needle) with your free hand.
- ✓ Align your sharp instrument so that you are pointing it at the far side of the mouse's face, at the base of the far ear or at the base of the far side of the mouth.
- ✓ Prick the freckle with the lancet. If using an 18 gauge needle, go in only up to the depth of the bevel.
- ✓ Quickly drop the sharp into the sharps container and pick up your collection tube.
- ✓ Collect 4-7 drops of blood (maximum amount depends on frequency of bleeding when in doubt, contact an DLAM veterinarian)
- ✓ Release the mouse into its cage when you have obtained your sample. Bleeding should cease immediately.

Lateral Saphenous Vein Blood Collection in Rodents

http://film.oslovet.norecopa.no/saphena/index.html

Maximum Blood Volume for Survival Collection in Lab Animals

The maximum amount of blood to be collected, as a survival procedure, from the following laboratory animals is 15% of the circulating blood volume. Frequency of collection should not exceed every other week. Hematocrit must be monitored and fluid replacement considered for protocols which require blood collection in larger volumes or at more frequent intervals.

Species	Total Blood Volume	15% Blood Volume
Rabbit	60 ml/kg b.w.	9.0 ml/kg b.w.
Rat	58 ml/kg b.w.	8.7 ml/kg b.w.
Mouse	78 ml/kg b.w.	11.7 ml/kg b.w.
Dog	90 ml/kg b.w.	13.5 ml/kg b.w.
Cat	66 ml/kg b.w.	9.9 ml/kg b.w.

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Training can be obtained through the DLAM by submitting a Special Request form to the Animal Resource Office.