

University of Notre Dame- Freimann Life Science Center

Procedure for Submandibular (Facial Vein) Blood Sampling

Purpose:

Retro-orbital blood sampling has been banned in Europe for several years. FLSC has evaluated alternatives to the retro-orbital route to promote the most humane procedures among the animal care staff and researchers.

Equipment:

Paper towels
Cotton ball(s)
Goldenrod Lancets (4 mm, 5mm or 5.5 mm) preferred **OR** 22g – 18g sterile needle
Eppendorf tube 1.5 ml
Heparinized hematocrit tube

Procedure:

1. Select either needle or lancet depending on the sample volume, sex and age of the mouse.
2. Restrain the mouse by scruffing the skin at the back and neck.
3. Identify the landmarks for the venipuncture. The submandibular vessel is found where lines drawn from the corner of the eye and the corner of the mouth intersect near the lateral/caudal aspect of the mandible. This coincides with a small hairless dimple found caudal to the corner of the mouth.
4. Insert the lancet or needle perpendicular to the skin to a depth of 4 – 5.5 mm.
5. Upon removal of the needle or lancet, blood will begin to flow.
6. Collect the blood in either a heparinized hematocrit tube (small sample) or allow the blood to collect into an eppendorf tube (large sample). For blood smears a single drop may be placed directly on the slide.
7. Release the scruff and the bleeding should stop. Ensure hemostasis by blotting with either a cotton ball or a paper towel before returning the mouse to the cage.

Technical Tips:

1. When using a needle the bevel should face toward the body of the mouse.
2. Select the size of the lancet or needle according to the sex and age of the mouse (these are approximations and may vary with strain)

4mm lancet/22g needle	3 wk – 4 wk old mice (under ~15 g)
5mm lancet/20g needle	female mice under 10wks (under ~20g)
	male mice under 6 wks (under ~20g)
	for single drop samples (smears)
5.5mm lancet/18g needle	female mice over 10 wks (over ~20g)
	male mice over 6 wks (over ~20g)
	large sample volumes

3. Alternating sides for repeated blood sampling is recommended if sample intervals are less than 5-7 days
4. When using the needle method, avoid insertion beyond a depth of 5.5mm as trauma to the muscles, nerves and vessels may cause excessive hemorrhage resulting in hypovolemia.
5. The most important part of this technique is the restraint. If scruffed too tightly blood supply and breathing may be restricted. To assist blood flow, position the mouse with the head lower than the heart. For better accuracy in puncturing the vessel, position the mouse in lateral recumbency.