

# Freimann Life Science Center

## Standard Operating Procedure for Saphenous Vein Blood Sampling in the Rat

### Purpose:

The saphenous vein in the rat is a readily accessible venipuncture site as an alternative to retro-orbital bleeding. In addition, anesthesia is not required to obtain a sample. This method of blood collection provides a sample size from 75 microliters to 225 microliters.

### Equipment:

Electric clippers	22g x 1" needles
Cotton balls	Cotton tipped applicators
Rodent DecapiCone®	Triple antibiotic ointment
Canvas gloves	Hematocrit tubes (Plain or Heparinized)
Clay sealant	Eppendorf tubes

### Procedure:

1. The rat is restrained in dorsal recumbency around the chest with the forelegs drawn up under the chin. The hindquarters will be presented to the person taking the sample. As an alternative to manual restraint, a plastic DecapiCone® with an opening cut level with the hind leg may be used. The rat is placed into the DecapiCone® and the hind leg pulled through the hole. The excess plastic can be gathered around the tail to prevent the rat from backing out.
2. The back leg is extended and the hair shaved from the inner surface of the leg from the hock to the groin.
3. Apply a small amount of triple antibiotic ointment to the shaved area using the cotton tipped applicator. Only apply enough to form a thin layer as a barrier between the skin and the blood.
4. Apply digital pressure at the groin to raise the femoral and saphenous veins. The vein crosses the leg at the ankle and wraps around below the knee joint.
5. Using the 22 g needle make a puncture by inserting the needle perpendicular to the vessel on the inner surface of the hind leg. The blood should immediately flow and form a drop on the skin.
6. Fill hematocrit tubes from the blood that pools on the surface of the skin. Up to 3 tubes can be collected.
7. Release leg and apply pressure to the puncture site to achieve hemostasis.
8. Return the animal to the cage after checking that bleeding has stopped.
9. DecapiCone® restrainers may be used for multiple rats and discarded when soiled or torn.

### Precautions:

1. When restraining manually, to avoid injury to personnel and animals, large or fractious rats can be lightly anesthetized using inhalation anesthesia (Isoflurane) in a bell jar.
2. Leather gloves may be substituted for canvas gloves if the rat is an adult and the person restraining feels it is advantageous.
3. Support the back of the rat with a gloved hand. Use the free hand to prevent the rat from twisting or bouncing by placing it over the abdomen.
4. Before returning the animals to their room, check again that hemostasis has been accomplished.
5. Blood samples can be spun in the hematocrit tube or transferred to an eppendorf tube to suit the needs of the protocol.