Curtin University Standard Operating Procedure

Blood Collection Techniques in Mice and Rats

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Aim/Purpose: To provide directions for the collection of blood from rodents using the tail vein or the lateral saphenous vein.

Definitions:

PPE: Personal Protective Equipment.

Technical Term 2: Definition

All employees have a duty of care (see Occupational Health and Safety) to ensure their own health and safety, and that of their fellow workers at all times.

ALL ANIMALS MUST BE HANDLED HUMANELY i.e. carefully and kindly.

Materials:

- Sterile 27-30 gauge needles for Mice
- Sterile 23-27 gauge needles for Rats
- Syringes
- Heating device
- Ethanol
- Gauze / Cotton Swabs
- Anaesthetic / Anaesthetic equipment if required
- Restraint if required.
- Clippers
- Collection tubes if required – ensure correct ones for blood / serum required.

- PPE

Calculations:

Weigh each animal before injection.

See the table below with the appropriate amount of the animals body weight in volume can be collected at any one time point.

Species (Weight)	Total Blood Volume (ml)	7.5% (ml)	10% (ml)	15% (ml)	20% (ml)
Mouse (25g)	1.8ml	0.1ml	0.2ml	0.3ml	0.4 ml
Rat (250g)	16ml	1.2ml	1.6ml	2.4ml	3.2ml

Total blood volume = approx. 6 % of total body weight.

Safe Acute Blood Sampling – acute blood sampling is the one-time removal of a large volume of blood or multiple small samples of blood over a short period of time (24 hours)

- 10-15% of circulating blood volume may be removed once every 3 weeks
- 1% of body weight can be collected every 3 weeks (or in total over a 24 hour period)

Chronic Sampling – chronic sampling is the frequent and repeated removal of small quantities of blood over a long period of time.

- The Rule of Thumb is 0.1% of body weight every day for 21 days (e.g. a 30gm mouse can have 0.03ml of blood collected every day for 21 days)
- The total volume of blood collected by chronic sampling is higher than acute, as the body continuously produces blood to replace that taken.

Procedure for Saphenous Vein Blood Sampling:

1. Choose the appropriate restrainer for the species
2. The animal is held in the restrainer head first so that only the rear legs and tail are free. The rear leg is then stretched out into a natural position.
3. The skin on the upper thigh is gently by firmly squeezed, using the same hand that is holding the restrainer.
4. Clip the hair over the thigh region. Swab the skin with a small amount of ethanol to help visualise the vein.
5. Locate the lateral saphenous vein
6. Using a 25-26 G needle or an animal lancet, puncture the vessel at a 90 degree angle at the most proximal (closest to the body) visible site.
7. Collect the sample into your collection tube ensuring you do not exceed the allotted blood volume loss.

8. Use a dry piece of swab or cotton wool to apply pressure to the puncture site while releasing pressure on the upper thigh until the bleeding stops.
9. Remove the mouse or rat from the restrainer and place it back in its cage
10. Monitor the animal for 5-10 mins to ensure the bleeding has stopped
11. For repeat samples, the scab may be brushed off with a dry piece of gauze or a new puncture site can be made distal to the previous site (towards the foot)

Blood Volumes:

Volumes as large as 5% of the circulating blood volume can be taken from the saphenous vein.

Procedure for Tail Vein Sampling Techniques:

1. Warm the mouse / rat
2. Place the mouse / rat in the appropriate restrainer. Rats may be better wrapped in a towel or laboratory coat.
3. Prep the tail by washing with water and then a wipe with ethanol/ chlorhexidine.
4. Visualise the tail vein on either side of the tail
5. Prick the vein perpendicular to the tail. The initial prick should be as near to the tip of the tail as possible. This will allow one to make repeated nicks if possible.
6. Allow the blood to flow into the tube by capillary action. Do not attempt to 'milk' the blood from the tail- this may cause tissue damage and contamination of the blood sample with tissue fluids.
7. When done collecting the blood, ensure pressure is put on the area to achieve haemostasis.
8. Place the animal back into its cage and observe for normal behaviour.

Blood volumes:

This doesn't give large amounts of blood easily – approximately 0.1-0.15ml in a mouse, and up to 2ml in warmed rats.

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Reviewed:

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