

LAB_020 Blood Collection – Facial Bleed (Sub-Mandibular) in Mice (Expiry: October 2028)

I. OBJECTIVE

To describe the standard facial vein blood collection procedure in mice and rats used across UQ research projects, also reflecting the procedure used to trainer workers across UQ by UQBR.

NB: The use of (*) indicates this statement is dependent on the facility procedures

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II. DEFINITIONS

Competent - “the consistent application of knowledge and skill to the standard of performance required regarding the care and use of animals. It embodies the ability to transfer and apply knowledge and skill to new situations and environments.”¹

III. COMMENTS/RECOMMENDATIONS

- Facial bleeds must be performed by appropriately trained personnel who have been deemed to be competent in the procedures.
- A new lancet should be used on each animal, this is to ensure a sharp lancet and to minimise contamination.
- The frequency and volume of blood collections will be affected by the lifetime experimental burden on the mouse and the health status of the mouse. For example, anaemia, tumours, kidney disease, liver disease or old age will reduce the resilience of the mouse to blood collection. Smaller volumes taken less frequently will be required.

Calculation of blood volume to collect

It is vital that the correct volume of blood to be collected is calculated. Blood collection volume is generally calculated as a % of total blood volume. This is the clinically relevant value. Given that the average blood volume of a mouse is 7% of body weight the following is an example calculation for a mouse weighing 20g:

Total blood volume is 7% of 20 g

$0.07 \times 20 = 1.4\text{ml}$

10% of total blood volume is $0.10 \times 1.4 = 0.14 \text{ ml}$ (or 140 μl)

(note: 1g of body weight = 1ml of blood)

Table 1. Recommended blood collection volumes based on mouse body weight (NHMRC 2008).

Body Weight (BW)	Total Blood Volume (TBV) [~7% of BW]	Minor Bleed (<7.5% of TBV)	Moderate Bleed (7.5-10% of TBV)	Major Bleed (10-15% of TBV)	Lancet Tip Size
Minimum Recovery period:		1 week	2 weeks	3 weeks	
18g	1.2mL	<90uL	90-120uL	120-180uL	Max 4mm
22g	1.5mL	<115uL	115-150uL	150-225uL	Max 4.5mm
26g	1.8mL	<140uL	140-180uL	180-270uL	Max 5mm
Attempt allowances	3 attempts per cheek.				
Procedure frequency	Dependant on bleed volume and minimum recovery period as advised in this table.				

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 THE UNIVERSITY OF QUEENSLAND AUSTRALIA CREATE CHANGE	UQ Animal Ethics Committee - Standard Operating Procedure LAB_020 Blood Collection – Facial Bleed (Sub-Mandibular) in Mice Institutional author: UQ Biological Resources AEC Reviewed & Approved: October 2025 SOP Expiry: October 2028	Version #4.0 Page 2 of 5
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IV. SAFETY AND COMPLIANCE

- The person undertaking this task must ensure all relevant approvals are in place, training has been undertaken and risk assessments have been performed. If unsure, consult your supervisor.
- Facility protocols should be followed.
- Possible risks include mouse bite injury, needle stick injury, spills, exposure to infectious agents, repetitive task musculoskeletal injury and psychosocial harm.

V. TRAINING CONSIDERATIONS

- All unsupervised animal blood collection must be performed by appropriately trained personnel who have been deemed to be competent in the procedure.
- Training facial vein blood collection must be undertaken on models or cadaver animals initially.
- Further training should be undertaken on animals under general anaesthesia.
- Note for UQBR Training purposes, a minor bleed (about 7% of blood volume) will be collected.
- For UQBR training purposes animals may be kept for monitoring. Adverse effects may take time to develop and can assist with the assessment of competency.

VI. EQUIPMENT

- PPE *
Minimum PPE is gloves and gown, additional PPE may be required based on facility or additional risk e.g. working with infectious animals.
- Disinfectant *
- Sharps Container
- Clinical waste bin
- Change station/Bio-safety cabinet *
- Blood collection tube or capillary tube
- Lancet 3-5mm, depending on size and age of the animal see table 1.

VII. PREPARATION

- Check AEC approvals to ensure that the correct procedure and personnel are approved for the planned work.
Deviations can occur between approved procedures listed versus what is planned with the animal – check that these match and that the relevant personnel are approved.
- Set up equipment items.
There should be no contamination of equipment during this process.
- Turn on Change station or Biosafety Cabinet. *
- Wipe surfaces with disinfectant.
Ensure equipment is operating as required.

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VIII. PROCEDURE

1. Check that the sample collection tube type is correct. *E.g. does it require anti-coagulant?*
2. Open the sample collection tube and prepare for blood collection. Ensure it is free of contaminants and is ready for blood to drip into.
3. Ensure you have the correct rodent for this procedure.
Check identification marks and ensure this matches the labelling on the collection tube.
4. Unless proficient in the technique, anaesthetise the mouse using gaseous anaesthetics (i.e. isoflurane).
Someone who is consistent and confident in extracting the required amount of blood in the first or occasionally (<10%) the second attempt may not need to anaesthetise the mouse as the level of stress should be minimal. During the training stage, individuals may be trained in fresh cadavers or anaesthetised animals, progressing to conscious animals depending on the trainee skill and confidence.
5. Restrain the animal as per **LAB_006 Handling and Restraint in Mice and Neonates** for mice and **LAB_039 Handling and Restraint in Rats and Neonates** for rats.
Sufficient restraint of the head without restricting blood flow is critical to achieve a successful bleed. A pinch grip (using thumb and forefinger on the opposing cheek) can be useful.

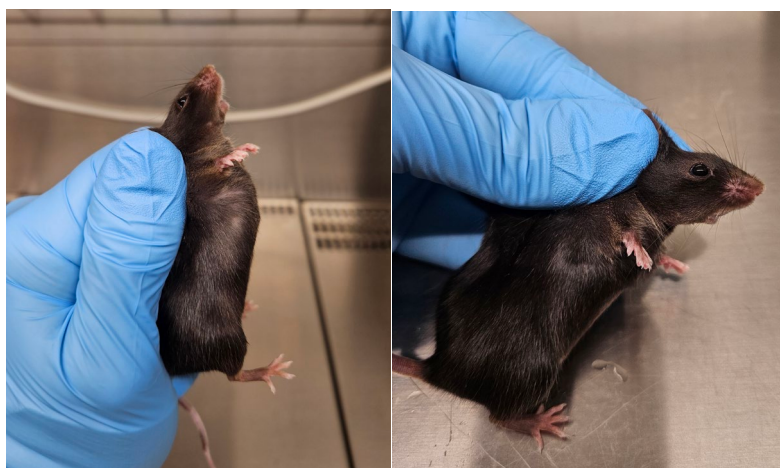


Figure 1. Appropriate restraints for this technique (UQBR 2019).

6. Position the animal above the sample collection tube in preparation for puncture.
Taking care not to contaminate the tube with urine/faeces etc.
7. Locate the facial vein puncture site by using the scent gland as a frame of reference or the temporal vein puncture site using the bulge of the cheek muscle and aiming for the caudal edge (toward the rear).
Alternatively, you can locate the temporal vein puncture site by drawing lines from the bottom jaw to the ear and the top of the eye to the shoulder. Where the lines intersect is the location of the puncture site.

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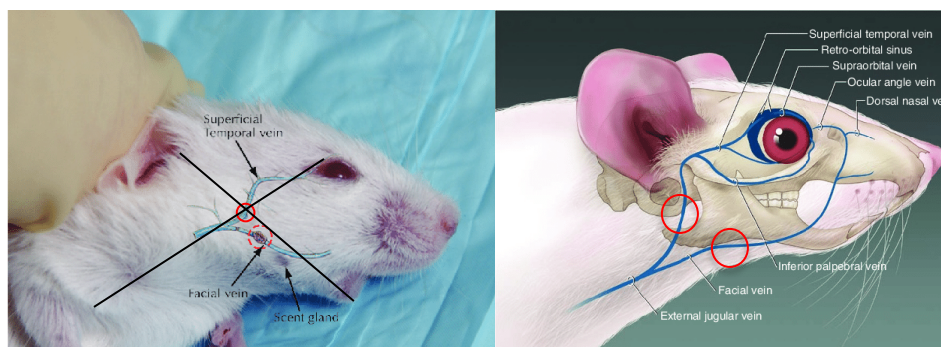


Figure 2. Puncture sites for this technique

8. Swiftly (<1s) puncture the vein with the lancet.

The lancet should be angled to puncture perpendicular (90 degrees) to the surface of the skin. Blood should drip from the site of puncture. If this does not occur this is commonly due to the puncture being too shallow, vein restriction, or the site being incorrect.

9. Collect blood sample into blood collection tube.

The total volume of blood collected must include all drops of blood, including those not collected/those on the tissue when stopping the bleeding. The exact quantity of blood that drips from the site can be difficult to control. Therefore, this method should not be used for more frequent sampling or methods that only require small volumes of blood. If insufficient blood is collected recheck the site and depth of puncture.

10. Apply pressure to the puncture site with a tissue or gauze to stop the bleeding.

Monitor the breathing to ensure the pressure is not restricting the airway.

11. Release rodent into the holding cage and continue to monitor its health.

Care of mouse after blood collection

- Monitor until the mouse is fully recovered from anaesthesia.
- Mice must be checked the following day for adverse effects. Record must be kept of this monitoring.
- Following the procedure, the mouse should return to normal movement and behaviour.
- Most animals will recover from this without further adverse effects if left undisturbed. In the rare case that bleeding re-commences, the animal should be restrained again and a tissue or gauze securely held to the site for 30-60s to encourage clotting. Ensure pressure is consistent and firm, but not hard.
- Refer to LAB_022 Veterinary Care Protocol for management of adverse effects:
 - Bleeding from the ear, nose or mouth.
 - Hematoma formation
 - Head tilt or circling

12. Place lancet into sharps container and close the sample collection tube.

The sample collection tube should be closed without contamination and stored appropriately (e.g. refrigerated if required by the research).

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- 13. Complete record keeping requirements – note the details of the procedure, date and initials. Log procedure on relevant AEC animal monitoring paperwork and the relevant research sample collection labelling/records.
Records need to be clear and legible on each record to allow others to read and understand.
- 14. Store the sample as required (e.g. refrigeration).
- 15. Repeat from step 1 for the next animal or if finished, pack and clean up equipment and space.

IX. BIBLIOGRAPHY

- 1. National Health and Medical Research Council (NHMRC) 2008, Guidelines to promote the wellbeing of animals used for scientific purpose <https://www.nhmrc.gov.au/about-us/publications/guidelines-promote-wellbeing-animals-used-scientific-purposes>

Version #	Reviewing AECs	AEC Review Date	Outcome
3.0	Molecular Biosciences AEC, Laboratory Biomedicine AEC, Anatomical Biosciences AEC, Health Sciences	March 2025	Extended to March 2026
4.0	Molecular Biosciences AEC, Laboratory Biomedicine AEC, Anatomical Biosciences AEC, Health Sciences	December 2024-October 2025	Approved

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