RESEARCH AND INNOVATION
ANIMAL RESOURCES AND CARE
VIRGINIA TECH

Standard Operating Procedure: Blood Collection in the Mouse, Intracardiac

These SOPs were developed by the Office of the University Veterinarian and reviewed by Virginia Tech IACUC to provide a reference and guidance to investigators during protocol preparation and IACUC reviewers during protocol review. They can be used as referenced descriptions for procedures on IACUC protocols.

However, it is the sole responsibility of the Principal Investigator to ensure that the referenced SOPs adequately cover and accurately represent procedures to be undertaken in any research project. Any modification to procedure as described in the SOP must be outlined in each IACUC protocol application (e.g. if the Principal Investigator plans to use a needle size that is not referenced in the SOP, simply state that alteration in the IACUC protocol itself).

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I. Procedure Summary and Goal

Describes procedures for the collection of blood from the heart as a terminal procedure in the mouse.

Considerations:

a. This is a non-survival (terminal) procedure.

b. This procedure must be performed under deep general anesthesia.

c. Please refer to the Guidelines for Injections in Rodents and Rabbits, Virginia Tech Office of the University Veterinarian for recommended volumes and needles sizes.

d. Sample volume obtainable – 0.5 to 1.0 ml (3-5% of body weight)

II. Personal Protective Equipment (PPE) and Hygiene

a. Ensure appropriate PPE is used to protect technician from accidental exposure to blood and other body fluids, such as:

   i. Gloves

   ii. Eye protection

   iii. Mask

   iv. Other PPE as required by protocol/facility

b. Hands should be washed and/or gloves changed between animals.

c. Promptly dispose of used sharps in the provided leak-proof, puncture resistant sharps container.

III. Supply List

a. General anesthetic - either alone or a combination of the following:

   i. Isoflurane – inhaled to effect, usually approximately 3%

   ii. 70% CO2 + 30% O2 gas – inhaled to effect

   iii. Ketamine (90-120mg/kg) + Xylazine (8-12mg/kg) combination

      1. Intramuscular, subcutaneous, or intraperitoneal injection

b. Syringes (1cc tuberculin or 3cc)

c. Needles (23 to 27 gauge; ⅝-1 inch)

IV. Detailed Procedure

a. Frequency

   i. Blood collection via cardiac puncture is a terminal procedure.

b. Anesthesia

   i. General anesthesia required

c. Procedure

i. Deeply anesthetize the animal with the selected anesthetic agent prior to sample collection procedures.

ii. Place the animal in dorsal recumbency once the animal has reached an appropriate plane of anesthesia (Figure 1).

iii. Attach appropriately sized needle to a syringe and insert bevel up at a 30-40° angle through the diaphragm, with syringe parallel to the midline of the mouse (Figure 2).

iv. Insert needle slightly left of and under the sternum, directed toward the animal's head. The needle can be angled slightly towards the left shoulder.

v. Retract the plunger slightly to create a vacuum inside the syringe and gently advance needle until blood flash appears in needle hub.

vi. Immobilize the needle and continue to aspirate until a sufficient amount of blood has been collected (Figure 3).

vii. Euthanize the animal immediately upon completion of blood collection.

1. Cervical dislocation

2. Bilateral thoracotomy

Figure 1. Correct Positioning

Figure 2. Insert Needle at 30-40°

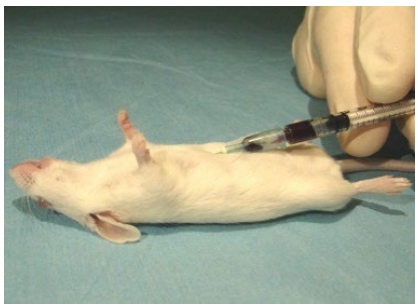

Figure 3. Aspirate Blood

V. Variations

a. Left lateral approach

i. Place the animal in right lateral recumbency

ii. Palpate the heart on the left lateral thoracic wall

1. At approximately the point of flexed elbows, between the 5th and 6th ribs

iii. Insert needle slowly, perpendicular to the body

b. Open approach

i. Place animal in dorsal recumbency

ii. Wet skin with 70% alcohol

iii. Make a V-shaped cut through the skin and abdominal wall about 1 cm caudal to the last rib

iv. Move abdominal organs aside

v. Insert the needle through the diaphragm and into the vena cava or heart

c. If blood stops flowing, rotate the needle or move it very slightly in or out.

VI. Potential Adverse Events, Mitigation, or Treatment

a. Distress due to restraint or blood loss

i. Perform secondary method of euthanasia

ii. Place back in gas anesthetic, use face mask for continuous delivery of gas anesthetic

VII. References

American Association of Laboratory Animal Science. Laboratory Animal Technician Training Manual. (Memphis, TN: Drumwright and Co, 2007)

Charles River Insourcing Solutions. Biomethodology in the Laboratory Mouse Charles River SOP 2405-3 - Dosing of Rodents– TGS and Discovery Services Charles River SOP 2577-2 - Blood Collection Methods for Use in Studies

Hawk, C.T., Leary, S.T., and Morris, T.H. Formulary for Laboratory Animals (3rd ed.). (Ames, Iowa: Blackwell Publishing, 2005) http://www.research.usf.edu/cm/docs/Formulary_for_Lab_Animals_3rd_ed.pdf

Hoof, J. Methods of Blood Collection in the Mouse. Lab Animal (NY). 29(10). (2000 Nov)

National Institutes of Health Animal Research Advisory Committee. Guidelines for Survival Bleeding in Mice & Rats. http://oacu.od.nih.gov/ARAC/survival.pdf

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