Research and Innovation Division

Research Animal Standard Operating Procedure

SOP# 24

Research Animal Standard Operating Procedures (SOP) must meet the following criteria:

1. Describe procedures or activities involving research animal(s) common to more than one research project.

2. Support the handling and or performance or undertaking of a procedure(s), involving an animal, in the same way on each occasion it is performed.

3. Describe a procedure or activity involving a research animal(s) undertaken by more than one person; and

4. Describe a procedure or activity involving a research animal(s) that will be undertaken in more than one location.

Name of Procedure	Blood collection – tail vein	
Species	Rat	
ACEC	**Reference**	SOP#24 – Mar 23 - Blood collection from the tail vein in the rat
	Author	Jenny Smart
	Version	1.5
	Date approved	24 March 2023
	Date for review	24 March 2026
	Procedure classification 1. Observation involving minor interference 2. Animal unconscious without recovery 3. Minor conscious intervention 4. Minor surgery with recovery 5. Major surgery with recovery 6. Minor physiological challenge 7. Major physiological challenge	3
Ethical considerations	1. Respect for animals must underpin all decisions and actions involving the care and use of animals for scientific purposes. 2. The procedure must be performed according to current best practice to support the wellbeing of the animal. 3. Persons performing this procedure must be competent in the procedure or be under the direct supervision of someone who is competent.	

Details

Site of Blood Collection

Tail vein

Description of procedure

1. MATERIALS

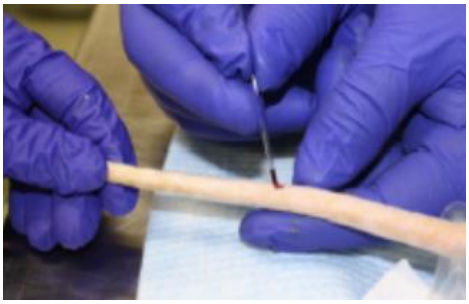
1.1 Sterile Butterfly Needle or standard hypodermic needle(23G-26G)
1.2 Blood collection device (e.g. Capillary tube)
1.3 Gauze
1.4 Antiseptic such as Chlorhexidine in 70% ethanol in water
1.5 Restraint device
1.6 Heat lamp (optional)
1.7 Weighing machine

2. PROCEDURES

2.1 Set up work surface with the above materials.
2.2 Weigh rat and calculate approximate blood volume and maximum blood collection volumes based on an average of50- 70 mls blood/ kg weight and maximum volumes as shown in section 3.
2.3 Place the rat in a restraint device such as a commercially available restraint tube or ‘rat bag” so that the tail of the rodent extends downward to the floor or anaesthetise the rat as per approved protocol.
2.4 The veins are located on the lateral surface (either side) of the tail. If the vein is not readily visible, place the rat under a heat lamp for a few minutes to cause the vein to dilate. Care must be taken not to overheat the rat or burn its ears. Place the lamp at 45-60cm away from the animal. Place your hand at the animal's level for at least 1 minute to determine if the lamp is too close to the animal. Placing the tail in a bowl of warm water (no warmer than 40°C) can also dilate the veins. Gently wash the side of the tail over the vein with gauze soaked with antiseptic.

1.1 **Collection**

2.5 :
(i) Occlude the vein by placing the base of the tail between your thumb and index finger or between your index finger and middle finger. This will cause the vein to dilate.
(ii) Insert a needle into the vein until a flash of blood is seen. Start as distal (far away) from the base of the body as possible as it is easier to successfully hit the vein lower in the tail. If you are initially unsuccessful, insert the needle directly above the last venipuncture site. The vein is very shallow so do not insert the needle at any angle to the tail.
(iii) Once the needle hub fills with blood, do not remove the needle from the tail. Insert the capillary tube(s) into the needle hub and allow gravity to fill the tubes.
(iv) Once the blood sample is collected, remove the needle from the tail and apply pressure to the venipuncture site to encourage hemostasis.
(v) Gently wash away any traces of blood before returning the rat to its cage.



NOTE: Discard all garbage into appropriate containers and clean the work area.

Maximum volume of blood will be collected?

(i) Single Bleed

Maximum removable volume should be no more than 10% of the animal's blood volume (see table below).

(ii) Multiple samplings

Maximum removable volume on a daily basis should be no more than 1% of the animal's blood volume (see table below).

Rat		
Body Weight	150 gms	250 gms
Estimated whole blood volume (ml/kg)	50-70	50-70
Blood volume (ml)	7.5- 10.5	12.5-17.5
Volume to be removed – single collection (10% of blood volume)	0.75-1.05 ml	1.25-1.75 ml
Volume to be removed – Multiple collections (1% of blood volume/day)	0.075-0.105 ml	0.125-0.175 ml

How will the animal be monitored for the effects of acute blood loss?

Signs to be monitored as indicative of acute blood loss in the rat include pale ears and feet, cold skin and extremities, restlessness, hyperventilation, and a subnormal body temperature.

Where multiple blood collections are to be performed:

Detail the total number of blood collections, and the time interval between each collection.

Repeated blood samples of 10% of total blood volume can be collected at interval of two- to three- week intervals or 1% of total blood volume on a daily basis. In these instances, the animal must be monitored for effects of chronic blood loss (see Section 6).

How will the animal be monitored for the effects of chronic blood loss?

Signs to be monitored as indicative of anaemia from chronic blood loss include:

- pale mucous membranes (conjunctiva or inside the mouth)
- pale tongue, gums, ears or footpads (if non-pigmented)
- increased respiratory rate when at rest (at the extreme level).

Additional monitoring should be performed when frequent blood collections are to be performed, with monitoring of the individual animal using its own baseline established at beginning of collection period. Monitoring parameters should include:

- packed cell volume
- haemoglobin level
- red cell count
- reticulocyte count

Peripheral blood smears can be examined in order to detect early changes associated with anaemia, for example, polychromasia of the red cells.

References

https://www.nc3rs.org.uk/rat-tail-vein-non-surgical

Removal of blood from laboratory mammals and birds: First report of the BVA/FRAME/RSPCA/UFAW Joint working group on refinement. Lab Anim (1993) 27, 1-22

ACEC Chair