

UBC Animal Care Guidelines

SOP: ACC-2014-Tech13 Collection of small amounts of blood from tail tip microsampling in rats.

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COLLECTION OF SMALL AMOUNTS OF BLOOD FROM TAIL TIP MICROSAMPLING IN RATS

Purpose:

This Standard Operating Procedure (SOP) describes the procedure for collecting small volume blood samples from the tip of the tail of rats. This procedure can be used to collect 5-40 ul of blood per time point. This SOP follows the UBC and CCAC guidelines for the collection of blood in laboratory rodents

Policy:

All persons must adhere to the guidelines of the Canadian Council on Animal Care and UBC Animal Care Committee policies and guidelines for the handling of and blood collection procedures from rats.

All animal users collecting blood from rodents must have successfully completed the UBC Animal Care Services (or equivalent) Rodent Biology and Husbandry course and must be listed on the approved Animal Care Committee Protocol.

Responsibility:

All researchers, Principal Investigators, students and technicians responsible for the humane handling and collection of blood samples from rats.

References:

1. Canadian Council on Animal Care (CCAC) guidelines (www.ccac.ca)
2. UBC Animal Care Committee; Policy on Acceptable Methods of Rodent Blood Withdrawal: [ANIMAL CARE POLICIES, SOPs & GUIDELINES | Office of Research Services](#)
3. National Centre for the Replacement, Refinement and Reduction of Animals in Research. **This video shows the procedure in mice
Video link: <http://www.nc3rs.org.uk/bloodsamplingmicrosite/page.asp?id=1335>

Calculations:

As a general rule most healthy adult rats have 60-70ml per kg of circulating blood volume.

1% (0.6 – 0.7 ml/kg) of this volume can be taken from healthy animals daily without deleterious effects.

10% (6-7 ml/kg) of this volume can be collected from health animals once every 2 weeks.

15% (9-10 ml/kg) of this volume can be collected from healthy animals once every 4 weeks.

**Collecting more than the recommended volumes or more frequently than recommended can lead to anemia and other clinical signs of illness and impact research results.

Materials

Appropriate Personal Protective Equipment depending on facility and biosafety requirements (lab coat, gloves, mask, bonnet, etc.)

25 G sterile hypodermic needle

“Sharps” container

70% Isopropyl alcohol

2x2 Gauze

Clean rat cage with wire lid

Paper towel

Towel for wrapping rat (tight weave, not terry towel so rats do not catch and rip toe nails; ensure the rat can breathe through the weave of the towel)

Safe heat source (if collecting samples >10 ul)

-Hot water circulation blanket, electric heating pad set on “low” to “medium”, infrared heater. ****Beware of heat lamps –if placed too close to the rat, burning of the ears is possible (place at least 24” away from rat).**

Appropriate blood collection supplies

i.e.: glucometer with blood collection strips, microhematocrit tubes, capillary tubes, etc.



Procedure

****If collecting samples <20 ul, warming the rat is typically not necessary but if needing to collect samples >20 ul, gently warming the rat by placing it in a clean empty cage lined with paper towel and placing the cage on a safe heat source for approximately 5 minutes will help dilate the blood vessels in the tail. Monitor closely for overheating (see adverse effects below). Do not leave rat unattended if being warmed. Alternately, the tail can be dipped into warm water (comfortably warm when you place your hand in it) for a few minutes.**

1. Get supplies ready for use.
 - a. Loosen 25G needle from cover carefully (do not poke yourself) and place close to where you can reach it. Leave needle partially in the cover to keep the tip sterile.
 - b. Moisten a 2x2 gauze with 70%
 - c. Get blood collection supplies ready (i.e: place glucose strip into glucometer or place capillary tubes in easy reach).

2. Gently remove the rat from the cage by grasping it at the base of the tail with one hand and scooping the body with the other hand being careful to support the entire body.
3. If rat is calm, one person can restrain rat by holding it against the person's body, along the person's. If another person is not available, the rat can be "burrito" wrapped.
 - a. Place rat on towel on flat surface.



- b. Put top of towel over rat's head and then hold rat through the towel behind its front legs to prevent the rat from backing out of the towel.



- c. Take the far side of towel and wrap it around the rat's body while still preventing rat from backing out of towel.



- d. Take the other side of the towel and wrap it around the rat's body – this will leave the tail exposed for the blood sampling.



- e. Tuck the wrapped rat into the crook of your arm. Ensure the rat can breathe and do not hold too tightly. Also ensure rat does not overheat.



4. While holding the tail with your non-dominant hand, wipe the distal (last) third of the tail with isopropyl alcohol gauze.
5. With your non-dominant hand, gently squeeze/stroke down the tail from the base of the tail to about 1 cm from the tip of the tail. The tip of the tail should become pinker than the rest of the tail. Hold gentle pressure on the tail with your non-dominant hand to occlude blood flow.
6. Place the tip of the tail on a flat area.
7. With dominant hand, perform the needle prick as close to the tail tip as possible (1-2 mm from end). The needle should be inserted perpendicular to the tail surface.



8. Collect the blood sample from the drop of blood that forms at the skin surface. Take care not to allow rat to pull its tail out of your grip. If more blood is needed, you can repeat the gentle squeezing/stroking of the tail from base to tip to encourage bleeding.



9. Release the pressure from your non-dominant hand and apply gentle pressure with gauze to tip of tail for approximately 10 seconds to stop blood flow.
10. Monitor tail tip for re-bleeding and if none seen, clean any blood of tail with gauze and warm water and return rat to its cage. If re-bleeding is seen, apply gentle pressure to tail tip again until bleeding has stopped. Take total amount of blood lost into account when planning next sampling time and amount.
11. Discard used needle into appropriate sharps container.
12. Note on cage card or procedure log the date and amount of blood sampled.
13. Check rat again approximately 15 minutes later to ensure no re-bleeding has occurred.
14. If repeated samples are needed the same day, it is usually possible to rub the previously sampled area with gauze and then gently stroking down the length of the tail to collect additional blood. If this does not work, the tail can be poked again on the opposite side of the first sampling.

Complications

1. Overheating if needing to warm rat
 - a. Extremities become very pink
 - b. Rat begins to lick itself and salivate
 - c. Rat moves away from heat source
 - d. Rat begins to “pant” – breathing deeply and rapidly
 - e. Rat may lie flat in cage and not move around normally
2. Too much blood loss causing anemia
3. Necrosis at tip of tail with repeated blood sampling
4. Discomfort resulting from insertion of needle (transient)
5. Stress resulting from restraint
6. Burns from heat lamps if placed too close to rat.