WASHINGTON STATE UNIVERSITY

Washington State University

Institutional Animal Care and Use Committee

Guideline #9: Blood Collection

Approval Date: 2/15/2023 (Replacing version 11/28/2022)

A. Purpose

This document is intended to provide guidance regarding safe volumes and common routes for blood collection from laboratory animals.

B. Guideline

The method of blood collection and volume collected can impact animal health and welfare. Repeated blood sample collection in mice, rats, hamsters, guinea pigs, small cats, and birds can be problematic because of their small body size. To prevent anemia, electrolyte imbalance, hypovolemic shock or other complications, the following guidelines should be followed:

1. The acceptable quantity and frequency of blood sampling is determined by the circulating blood volume and the red blood cell (RBC) turnover rate.
2. For optimal health, blood draws should be limited to the lower end of the range. Maximum blood volumes should be taken only from healthy animals.
3. The approximate circulating blood volume of an animal is 55-70 ml/kg. A blood volume estimate for a single species may not reflect differences among individual breeds or variations due to age, size, or illness. Older or obese animals can have a circulating blood volume that is up to 15% lower than the normal range.
4. Of the circulating blood volume, approximately 10% of the total volume can be safely removed every 2 to 4 weeks, 7.5% every 7 days, and 1% every 24 hours. The same total amount of blood can also be removed as multiple quantities over a 24-hour period. See **Table 1** for estimated blood volume for individual species. **Table 2** includes collection volumes for mice and rats of various weights. **Table 3** lists possible blood collection sites for multiple species.
5. With provision of replacement fluids (0.9% saline, Lactated Ringer's solution) equaling the volume of blood removed, up to 15% of circulating blood volume

may be collected at one time. This would require a 4-week recovery period before additional blood draws.

6. **When an increase of blood volume to be collected is requested for an IACUC approved protocol, anything over 10% every 2 weeks would need to go through a regular amendment process and would not qualify for a Veterinary Verification Consultation (VVC) according to WSU IACUC Policy #24.**
7. Although blood *volume* is rapidly restored in an animal after blood collection, the rest periods described above are needed for blood *constituents* (e.g., red blood cells, platelets, clotting factors) to be regenerated by the body. Hemostasis after collection can be achieved by using a silver nitrate stick, Kwik-Stop powder or by applying a gauze sponge over the site with gentle pressure until bleeding stops.

Table 1: Circulating Blood Volume & Examples of Maximum Collection Volumes Calculated for Survival Procedures.

Species	Circulating Blood Volume (mL/kg)	Average Adult Body Weight	Blood Volume	Maximum Collection Volume for Survival Procedures (10% of circulating blood volume)
Mouse	77-80	25 g	2 mL	0.2 mL or 200 µL
Rat	50-70	300 g	18 mL	1.8 mL
Hamster	78	85-150 g	9.2 mL	0.9 mL
Gerbil	67	45-130 g	5.9 mL	0.59 mL
Guinea pig	67-92	700-1200 g	75 mL	7.5 mL
Rabbit	57-65	2.5 kg	143 mL	14 mL
Cat	47-60	4 kg	214 mL	21 mL
Dog (beagle)	79-90	12 kg	1 L	100 mL
Ferret	75	1 kg	75 mL	7.5 mL
Pig (large)	65	110 kg	7.15 L	715 mL
Sheep	60	60 kg	3.6 L	360 mL
Goat	70	45 kg	3.15 L	315 mL
Cattle	60	520 kg	31.2 L	3.12 L
Horse	75	400 kg	30 L	3 L
Chicken	60	2.5 kg	150 mL	15 mL

Table 2: Approximate Blood Sample Volume Ranges and Safe Frequency of Collection for Mice and Rats.

Body Weight (g)	Total Blood Volume (mL)	1% (mL)	7.5% (mL)	10% (mL)	15% (mL)*
<u>Mouse</u> based on mean 78 mL/kg (blood volume/body weight)					
20	1.56	0.02	0.12	0.16	0.23
25	1.95	0.02	0.15	0.20	0.29
30	2.34	0.02	0.18	0.23	0.35
35	2.73	0.03	0.20	0.27	0.41
40	3.12	0.03	0.23	0.31	0.47
<u>Rat</u> based on mean 60 mL/kg (blood volume/body weight)					
125	7.50	0.08	0.56	0.75	1.13
150	9.00	0.09	0.68	0.90	1.35
200	12.00	0.12	0.90	1.20	1.80
250	15.00	0.15	1.13	1.50	2.25
300	18.00	0.18	1.35	1.80	2.70
350	21.00	0.21	1.58	2.10	3.15
Frequency of Collection		Every 24 hours	Every 7 days	Every 2 weeks	Every 4 weeks

*With provision of replacement fluids. See item 6 under Guideline.

Table 3: Blood collection sites for multiple species.

Species	**Site**
Mouse	Saphenous, tail veins or arteries, retro-orbital (requires anesthesia, less invasive methods preferred), submandibular and submental, **tail snip (requires anesthesia for rodents 21 days of age or older, less invasive methods preferred)
Rat	Saphenous, tail veins or arteries, pedal and jugular veins, **tail snip (requires anesthesia for rodents 21 days of age or older, less invasive methods preferred)
Hamster	Saphenous and jugular veins
Guinea pig	Saphenous, tarsal and jugular veins, vena cava
Rabbit	Ear vein or artery and jugular veins

Ferret	Jugular, cephalic and saphenous veins, cranial vena cava
Cat	Jugular, cephalic and saphenous veins
Dog	Saphenous, cephalic and jugular veins
Pig	Ear, saphenous, tail, cephalic, femoral and mammary veins, vena
Sheep & Goat	Jugular and cephalic veins
Cattle	Jugular veins
Horse	Jugular veins
Chicken	Brachial and jugular veins

**Tail Snip: Under certain circumstances, tail snip under anesthesia may be used as a collection method for small amounts of blood (less than 0.2 ml) in mice and rats only. Due to the potential for pain and permanent damage to the tail, it is highly recommended to avoid use of this method in favor of less invasive methods.

- *IACUC Approval for Use* – Justification for use and technique **must** be described in the ASAF and reviewed/approved by the IACUC prior to use.
- *Procedure Guidance* – Local or general anesthesia is required for use of this method in rodents 21 days of age or older unless adequately justified within the ASAF and approved by the IACUC. The fleshy tip of the tail may only be snipped **once** (less than 1mm) with a sterile blade at an angle perpendicular to the tail to avoid removal of bone. If performing serial collections, the tail **must not** be snipped again; however, subsequent collections are possible by gently removing the clot/scab for each sample. After collection, blood flow should be stopped by applying pressure for >45 seconds with a gauze pad or use of clotting agents (e.g., Kwik-stop).
- This method is not appropriate for animals that previously underwent snip for genotyping as it would require the snip to be taken further up the tail, increasing potential for removing bone.

If you have any questions about this guideline, need training in one of the above methods, or need the information regarding a species not listed above, please call the Office of the Campus Veterinarian at 509-335-6246 or email at or.ocv.alert@wsu.edu.

C. References

1. Diehl K, et al. 2001, A Good Practice Guide to the Administration of Substances and Removal of Blood, Including Routes and Volumes, J Appl Toxicol 21:15-23.
2. Iwarsson K, Lindberg L, Waller T, 1994, Common non-surgical techniques and procedures. Chapter 16. Svensen P, Hau J (eds). In: Handbook of Laboratory Animal Science. Volume 1. CRC Press, Inc. Boca Raton, FL.
3. National Research Council, 2011. Guide for the Care and Use of Laboratory Animals. 8th Edition. The National Academies Press, Washington, DC
4. S. Parasuraman, R. Raveendran, R. Kesavan, 2010. Blood Collection in Small Laboratory Animals. National Library of Medicine, PubMed Central, Bethesda, MD. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3043327/