

General guidelines:

- The acceptable quantity and frequency of blood sampling is determined by the circulating blood volume and the red blood cell (RBC) turnover rate. Excessive blood collection may result in hypovolemic shock, physiological stress and even death.
- 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 (1).
- The approximate blood volume of a mouse is 77-80 $\mu\text{l/g}$. **Single sample:** Without fluid replacement, the maximum blood volume which can be safely removed for a one-time sample is 10% of the total blood volume or 7.7-8 $\mu\text{l/g}$. With fluid replacement, up to 15% of the total blood volume or 12 $\mu\text{l/g}$ can be removed. Fluid replacement should be warmed and given subcutaneously (1) following blood collection. **Multiple samples:** If it is necessary to take multiple samples, smaller blood volumes should be drawn. The maximum blood volume that may be drawn per week is no more than 7.5% of the total blood volume. If sampling will occur every 2 weeks, up to 10% of the total blood volume may be drawn or 8 $\mu\text{l/g}$ (4). For a 25 g mouse, this is equivalent to 200 μl every 2 weeks. For repeated blood collection, fluid replacement does not substitute for a larger blood volume or more frequent blood collection.

Example for a 25 g mouse

	Without fluid supplementation	With fluid supplementation
Single Sample	193-200 μl	300 μl
Multiple Sample	145-150 μl per week	200 μl every 2 weeks

For different weights, please refer to: [NIH Office of Animal Care and Use Guidelines for Blood Collection in Mice and Rats](#)

> Please note these are general recommendations and there may be strain specific variations from these guidelines should be described in your IACUC protocol.

- **Exsanguination:** Approximately half of the total blood volume can be collected at exsanguination and must be performed under general anesthesia as a terminal procedure. This is equivalent to 40 $\mu\text{l/g}$ or approximately 1 ml for a 25 g mouse (1).
- Take into account the total blood volume yielded from the chosen blood collection technique when calculating frequency and volume of blood collection.

- If you are not experienced in blood collection technique and would like training contact: trainerIACUC@ucsf.edu

Collection Site*	Advantages	Disadvantages
Submandibular Sampling	<ul style="list-style-type: none"> • Preferred blood collection method • Maximum allowable sample volume with minimal trauma 	<ul style="list-style-type: none"> • Must be securely restrained • Anesthesia recommended • Yields a large sample so should not be used for frequent small sampling
Submental Sampling	<ul style="list-style-type: none"> • Minimal trauma • Multiple samples can be taken by alternating sites • May be easier due to direct visibility of the vessel 	<ul style="list-style-type: none"> • Anesthesia is required • Requires some specialized training
Tail Nick or Ventral Artery Sampling	<ul style="list-style-type: none"> • Anesthesia not required • Multiple samples can be taken • Vein is easily accessed 	<ul style="list-style-type: none"> • Must be securely restrained • Yields only small quantities • Some specialized equipment needed
Retro-orbital Sinus	<ul style="list-style-type: none"> • Yields a greater volume of blood 	<ul style="list-style-type: none"> • Anesthesia is required • Is a discouraged technique • Risk of injury to the eye and surround structures
Saphenous Sampling (medial or lateral approach)	<ul style="list-style-type: none"> • Anesthesia not required • Excellent technique for serial blood sampling • Moderate volume of blood can be collected 	<ul style="list-style-type: none"> • Requires some specialized training • Specialized equipment required
Cardiac Puncture	<ul style="list-style-type: none"> • Maximum volume of blood can be collected 	<ul style="list-style-type: none"> • Requires deep anesthesia. • Non-survival procedure only

*Refer to specific blood collection technique SOPs for more information.

References:

1. http://jaxmice.jax.org/faq/withdrawingblood_amounts.html
2. Mitruka BM, Rawnsley HM. 1981. Clinical, biochemical and hematological reference values in normal experimental animals and normal humans. New York: Masson Publishing; 413 p.
3. Harkness JE, Wagner JE. 1989. Biology and husbandry. In: Harkness JE, Wagner JE, editors. The biology and medicine of rabbits and rodents, 3rd ed. Philadelphia: Lea & Febiger.

4. McGill MW, Rowan AN. 1989. Biological effects of blood loss: implications for sampling volumes and techniques. *ILAR News* 31:5-20
5. Guidelines for the Survival Bleeding of Mice and Rats. 2010: oacu.od.nih.gov/ARAC/documents/Rodent_Bleeding.pdf
6. Regan RD, Fenyk-Melody JE, Tran SM, Chen G, Stocking KL. 2016: Comparison of Submental Blood Collection with the Retroorbital and Submandibular Methods in Mice (*Mus musculus*). *J Am Assoc Lab Anim Sci*. 2016; 55(5):570-6.