Video Article

Drawing Blood from Rats through the Saphenous Vein and by Cardiac Puncture

Christine Beeton, Adriana Garcia, K. George Chandy
Department of Physiology and Biophysics, University of California, Irvine

URL: <http://www.jove.com/index/Details.stp?ID=266>

DOI: 10.3791/266

Citation: Beeton C., Garcia A., Chandy K.G. (2007). Drawing Blood from Rats through the Saphenous Vein and by Cardiac Puncture. JoVE. 7. <http://www.jove.com/index/Details.stp?ID=266>, doi: 10.3791/266

Abstract

Drawing blood from rodents is necessary for a large number of both in vitro and in vivo studies. Sites of blood draws are numerous in rodents: retro-orbital sinus, jugular vein, maxillary vein, saphenous vein, heart. Each technique has its advantages and disadvantages, and some are not approved any more in some countries (e.g., retro-orbital draws in Holland). A discussion of different techniques for drawing blood are available ¹⁻³. Here, we present two techniques for drawing blood from rats, each with its specific applications.

Blood draw from the saphenous vein, provided it is done properly, induces minimal distress in animals and does not require anesthesia. This technique allows repeated draws of small amounts of blood, such as needed for pharmacokinetic studies ^{4,5}, determining plasma chemistry, or blood counts ⁶.

Cardiac puncture allows the collection of large amounts of blood from a single animal (up to 10 ml of blood can be drawn from a 150 g rat). This technique is therefore very useful as a terminal procedure when drawing blood from the saphenous would not provide a large enough sample. We use cardiac puncture when we need sufficient amounts of serum from a specific strain of rats to grow T lymphocyte lines in vitro ⁴⁻⁹.

Protocol

Drawing Blood from Rats through the Saphenous Vein and by Cardiac Puncture

Note: All procedures must be approved by your institute's animal care and use committee.

1. Blood draw from the saphenous vein

1. This procedure is performed without anesthesia and requires two persons, one who handles the rat, and one who performs the draw.
2. Make a cone out of a towel or hold the rat with rodent handling gloves, leaving one hind limb exposed.
3. Shave the back of the leg with an electric trimmer until the saphenous vein is visible. Shave a large enough area, so that no hair will come in contact with the puncture site. Use a small amount of non-fragrant hand lotion to keep the non-shaved hair away from the puncture site.
4. Make a compression point at the base of the leg to make the saphenous vein bulge out (similar to using a tourniquet when drawing blood from a human or a large animal). Puncture the vein using a 20G needle and scoop the blood as it comes out using a microvette. Pumping with the leg will help draw more blood. When you have collected enough blood, hold a clean compress on the puncture site to stop bleeding.

2. Blood draw by cardiac puncture

1. This procedure requires anesthesia and is terminal. Animals must immediately be euthanized at the end of a cardiac puncture.
2. Prepare a 5 ml syringe with a 23G1 needle.
3. Deeply anesthetize the rat and check for anesthesia by lack of spontaneous movement, slow breathing rate, and lack of response to stimuli (such as pinching a toe). To ensure prolonged anesthesia, place a syringe barrel containing paper towel soaked in volatile anesthetic on the rat's nose during the procedure.
4. Place the rat on its back, facing away from you.
5. If you are right-handed, place your left index finger at the level of the lowest ribs, without applying any pressure. The heart will be located ~ 1 cm above this point, slightly to the right.
6. Holding your syringe at a 45-degree angle, insert the needle between two ribs and watch for a drop of blood to come into the needle. This is an indication that you are in the heart. Without moving your syringe, pull on the plunger to fill the syringe. Once the syringe is full, carefully disconnect it from the needle and empty it into a tube. The syringe can then be re-attached to the needle for drawing more blood. It should be possible to draw 5-10 ml of blood from a 120-180 g rat.
7. Immediately euthanize the rat.

Discussion

Drawing blood from the saphenous vein is a convenient way to get small amounts of blood without anesthesia. If repeated sampling is required, check regulations to ensure you do not draw too much blood from one rat.

Drawing blood by cardiac puncture is a convenient way to get large amounts of blood but this is a terminal procedure. The animal must be euthanized at the end of the blood draw.

These techniques (as all techniques on live animals) should be first done in the presence of a veterinarian or a veterinary technician who will be able to assess that animals are handled properly and are not subjected to unnecessary pain and/or distress.

References

1. Van Herck, H., Baumans, V., Brandt, C.J.W.M., Boere, H.A.G., Hesp, A.P.M., Van Lith, H.A., Schurink, M., Beynen, A.C. Blood sampling from the retro-orbital plexus, the saphenous vein and the tail vein in rats: comparative effects on selected behavioural and blood variables. *Laboratory Animals*. 35, 131-139 (2001).
2. Luzzi, M., Skoumbourdis, E., Baumans, V., Conte, A., Sherwin, C., Kerwin, A., Lang, T., Morton, D., Barley, J., Moreau, E., Weilenmann, R.F., Reinhardt, V. Collecting blood from rodents: a discussion by the laboratory animal refinement and enrichment forum. *Animal Technology and Welfare*. 4, 99-102 (2005).
3. Angelow, O., Schroer, R.A., Heft, S., James, V.C., Noble, J. A comparison of two methods of bleeding rats: the venous plexus of the eye versus the vena sublingualis. *Journal of Applied Toxicology*. 4: 258-260 (2006).
4. Beeton C., Wulff H., Barbaria J., Clot-Faybesse O., Pennington M., Bernard D., Cahalan M.D., Chandy K.G., Beraud E. Selective blockade of T lymphocyte K⁺ channels ameliorates experimental autoimmune encephalomyelitis, a model for multiple sclerosis. *Proc. Natl. Acad. Sci. USA*. 98:13942-13947 (2001).
5. Beeton C., Pennington M.W., Wulff, H., Singh S., Nugent D., Crossley G., Khaytin I., Chen C.Y., Calabresi P.A., Chandy K.G. Targeting effector memory T cells with a selective peptide inhibitor of Kv1.3 channels for therapy of autoimmune diseases. *Mol. Pharmacol*. 67:1369-1381 (2005)
6. Beeton C., Wulff H., Standifer N.E., Azam P., Mullen K.M., Pennington M.W., Kolski-Andreaco A., Wei E., Grino A., Counts D.R., Wang P.H., LeeHealey C.J., Andrews B.S., Sankaranarayanan A., Homerick D., Roeck W.W., Tehranzadeh J., Stanhope K.L., Zimin P., Havel P.J., Griffey S., Knaus H.G., Nepom G.T., Gutman G.A., Calabresi P.A., Chandy K.G. Kv1.3 channels are a therapeutic target for T cell mediated autoimmune diseases. *Proc. Natl. Acad. Sci. USA*. 103:17414-17419 (2006).
7. Beeton C., Barbaria J., Devaux J., Benoliel A.-M., Gola M., Sabatier J.-M., Bernard D., Crest M., Beraud E. Selective blocking of voltage-gated K⁺ channels treats experimental autoimmune encephalomyelitis and inhibits T-cell activation. *J. Immunol*. 166:936-944 (2001).
8. Devaux J., Forni C., Beeton C., Barbaria J., Beraud E., Gola M., Crest M. Myelin basic protein-reactive T cells induce conduction failure in vivo but not in vitro. *Neuroreport* 14:317-320 (2003).
9. Beeton C., Chandy K.G. Induction and monitoring of adoptive delayed type hypersensitivity in rats, *Journal of Visualized Experiments*, 8, <http://www.jove.com/index/Details.stp?ID=325> (2007).