Jugular Vein Technique for Serial Blood Sampling and Intravenous Injection in the Rat (37402)

WILLIAM A. PHILLIPS, WALTER W. STAFFORD, AND JOHN STUUT, JR.
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Diabetes and Atherosclerosis Research, The Upjohn Company, Kalamazoo, Michigan 49001

The rat jugular vein affords a suitable site for obtaining uncontaminated sequential blood samples and for intravenous injections. Griffith and Jeffers (1), Holland *et al.* (2), Renaud (3) and others have reported methods using this site. Generally, these investigators entered the exposed vein after skin incision of the anesthetized animal. Renaud (3) suggested that blood collections and injections can be made directly through depilated skin when skin incision is contraindicated. In contrast, the procedure described in this report requires no prior preparation of the rat, which can be secured and released rapidly from a clamp-type restraining board. The plastic syringes used for blood collections are utilized in processing the samples after the rats have been bled.

Methods. The method we have developed requires a wooden restraining board 10 in. wide and 12 in. in length, supported by 4 legs 1½ in. in height for ease of handling. The board has 4 horizontal slits to permit adjustment of distances between common clip-type clamps (Esterbrook No. 20 ball bearing clips) placed at each corner. The metal ends of the clamps are covered with latex tubing held with adhesive tape to protect the legs of the animal. The same board can be used for rats weighing 100 to 600 g by changing the position of the clamps held by bolts and winged nuts.

The rat is secured on its back to the restraining board by the legs in the clamps as shown in Fig. 1. The forelegs are inserted first with the head forward. The board is turned 180° and twine is placed on the upper incisors. The head is drawn and held firmly to the left or right for entrance into either jugular vein. A 23 gauge ¾ in. disposable needle fitted to a 1 ml B-D tuberculin plastic syringe (sterile plastipak syringes and needles; Becton, Dickinson and Co.) is inserted bevel up in the angle made by the neck and shoulder in a cephalocaudal direction as shown. Entrance into the vein is cephalad to the point at which it disappears between the pectoralis muscle and clavicle. Generally, the bevel will be in the lumen of the vessel when about ½ the length of the needle has penetrated the skin. Insertion of the needle through muscle aids in stabilizing the needle in the vein. The plunger can be withdrawn rapidly to collect the desired amount of free-flowing blood without collapsing the vessel. Plasma samples may be obtained by treating the syringe with an appropriate anticoagulant. After removal of the needle, digital pressure for 10–20 sec over a cotton pad applied to the injection site prevents further bleeding and formation of a hematoma.

During the blood collection period samples are collated by inserting the needles, still attached to the filled syringes, into an 8 in. cork ring next to appropriate labels. To process samples the needles are broken off at the hub, by bending back and forth, Duco cement applied at that point, and identifying labels attached to the hubs. When the cement has dried (approx 15 min), the syringes are cut with aviation-type sheet metal snips immediately below the plunger, then centrifuged. Serum or plasma samples can be removed with an Eppendorf pipet, or a similar device, for analyses.

Results and Discussion. The procedure described represents a safe and rapid method

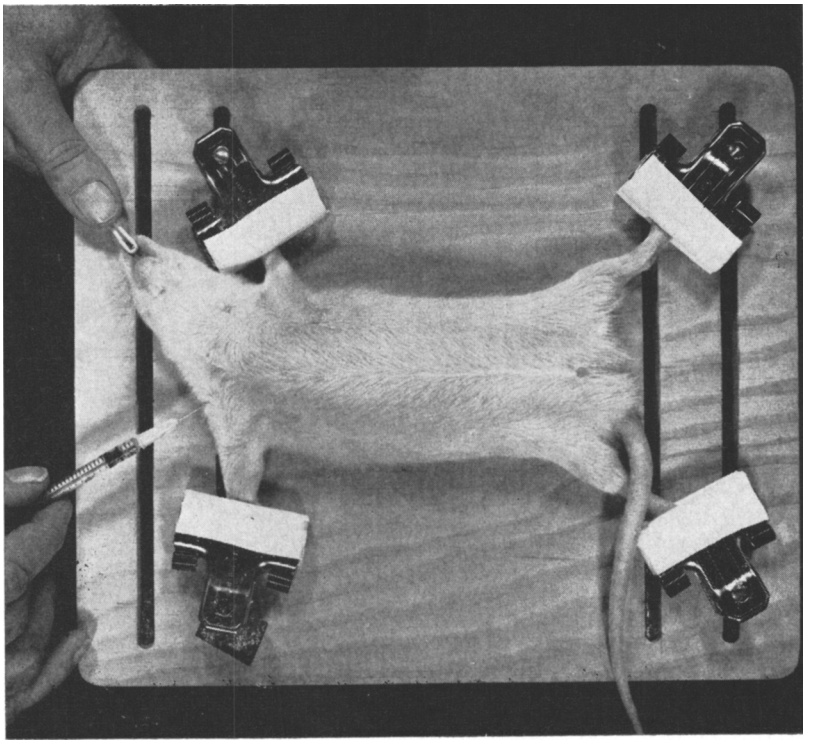

FIG. 1.

for intravenous infusions or obtaining repetitive blood samples from the same rat. A skilled technician can easily bleed 40 rats in 1 hr. Generally, we remove about 0.25 to 0.6 ml blood, although larger quantities can be obtained. Rarely does an animal die because of the procedure. Without anesthesia, rats will emit squeals of distress during the brief restraining period which can be avoided by use of light ether or other appropriate anesthetics. However, since anesthesia can also influence metabolic processes (4), its use depends upon the blood parameter to be measured. In our hands, weekly determinations of serum cholesterol concentrations using unanesthetized animals have been remarkably consistent for each rat during periods as long as 7 wk. Rats bled 1 or 2 times a week appear to be unaffected by the procedure. In acute studies as many as 7 blood samples, 0.25 ml each, have been obtained during a 24 hr period. As an example of the possible frequency of bleeding, Table I shows results obtained from a single rat during an acute drug absorption study. In this test, with the aid of one technician, 40 rats were bled at each of the time periods indicated. We have not run tests on hematopoiesis, but we have noted grossly a decreased hematocrit after frequent bleedings. We have never observed manifestations

TABLE I. Sequential Change in Serum Radioactivity in a Single Rat After Oral Administration of ^{3}H(G)-Digitoxin.

Hr	dpm/0.1 ml[a]
0.5	1317
1	1853
2	1961
4	1513
8	896
11	613
21	415
27	325

[a] Determinations made in Model 3375 Packard liquid scintillation spectrometer with fluid containing 8.4% Beckman Bio-Solv BBS-3, 0.4% PPO and 0.005% POPOP in toluene.

of septicemia.

Summary. We have described a technique for rapid serial blood sampling and intravenous injections via the jugular vein in the rat. The animal, without any prior preparation, is restrained on a specially designed board. Entrance into the vein is made cephalad to the pectoralis muscle. A trained technician can bleed 40 rats weighing 100 to 600 g in 1 hr. Considerable time is saved by using plastic syringes to collect the blood so that no transfers of samples are necessary for processing.

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2. Holland, L. M., Drake, G. A., London, J. E., and Wilson, J. S., Lab. Anim. Sci. **21**, 913 (1971).

3. Renaud, S., Lab. Anim. Care **19**, 664 (1969).

4. Bunker, J. P., and Vandam, L. D., eds., Pharmacol. Rev. **17**, 183 (1965).

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