

# A procedure for rabbit blood serial collection

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## ABSTRACT

Blood serial collection is required for many applied reproduction and nutrition trials with rabbits. The experimental design of these researches very often includes repeated samplings of large volumes of blood for metabolites and hormones analysis. Our proposed procedure has been tested in many surveys with success. It is based on a blood collection by aspiration, featuring a sterile butterfly cannula with a vacuum tube. By this procedure, the use of syringes is avoided. As a matter of fact when a syringe is used, excessive suction frequently causes the rabbit blood vessel collapse. Furthermore a rough handling of blood, such as excessive suction or the forcing of clotted blood from a syringe, will cause hemolysis: this can interfere with the serum or plasma fractions needed. Blood collection from ear vessels is a technique commonly used with rabbits. The central ear artery is recommended for collecting larger volumes of blood (over 5ml). 10ml of blood/kg body weight can be collected in this manner but the rabbit must be carefully restrained and hematomas must be prevented by direct pressure. Removal of more than 10% of blood volume may result in hypovolemic shock, a potentially fatal reaction.

**Materials.** For local anesthesia EMLA Cream (Lidocaine 2.5% + Prilocaine 2.5%) is a topically applied local anesthetic that is very useful in removing sensation from the venipuncture site.

**Blood collection set:** 1) Butterfly cannula (23 gauge needle for small blood samples and 21 gauge for large samples). 2) Vacutainer blood collection tube and vacutainer holder. 3) Luer adapter (which is designed to allow the attachment of the butterfly to holders using a single venipuncture). **Procedure.** a) Remove the rabbit from its cage. Proper technique in grabbing and carrying a rabbit is important to prevent vertebral fractures. Grab the rabbit's skin in the shoulder region and place its head underneath your arm. Support the rabbit's legs with the hand of the arm that the head is tucked under. Alternatively, grab the rabbit's skin with both hands (over both the shoulder and the rump region). b) Apply about one thumb size of anesthetic cream on the site selected for blood collection, and leave the cream in contact with skin for 5 minutes c) Check the ear artery if visible and if doesn't dilate, tapping it gently (or flicking it) may help. For vessels dilation the use of topical irritants such as xylene is not recommended. Xylene tends to cause leukocytosis and if it comes in contact with blood, will cause hemolysis. Adequate dilation of vessels can generally be achieved by use of a heat lamp, alcohol or gentle massaging of the ear. d) Only when the blood vessel is clearly visible, with the needle bevel up, parallel to, and alongside the artery, insert the needle quickly under the skin and then into the artery. The needle has to be pointed towards the base of the ear and the insertion into the skin and artery can be performed in one complete motion. e) If the blood vessel has been correctly entered, blood will flow into the tubing of the butterfly cannula. f) Push the vacutainer tube all the way into the holder; vacuum is broken, and blood flows freely being drawn from the artery into the tube. g) Once the blood sample has been taken, the needle must be carefully and quickly removed. When the desired amount is collected, gently hold gauze over needle entry point and remove the needle. h) Hemostasis should be achieved applying pressure to the injected area using thumb and a piece of cotton. Keep pressure on the area for 60-90 seconds, or until blood stops flowing. i) Observe the rabbit for at least another 30 seconds to insure that bleeding has stopped. l) Return rabbit to cage, and check again in 10-15 minutes.