

## METHODS OF OBTAINING EFFLUENT BLOOD FROM THE ADRENAL GLAND IN CHRONIC EXPERIMENTAL CONDITIONS

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Changes in the functional state of the adrenals may be demonstrated by many indirect tests, but these tests are inadequate for the more detailed study of their working.

A more complete picture of the function of the adrenals may be obtained by the use of methods of direct determination of the steroid hormones in the peripheral blood of animals. In this case, however, it is essential to bear in mind that the corticosteroids, secreted into the adrenal vein, are utilized quickly by the tissues and, on the other hand, that various metabolites may enter the peripheral blood. Moreover, the low concentration of the hormones in the peripheral blood complicates the estimation of the corticoids secreted by the adrenal gland in small quantities.

The fullest picture of the secretory activity of the gland may be obtained by determining the hormones in the outflowing venous blood. In the majority of investigations carried out in this direction, blood flowing from the adrenals was collected in acute experimental conditions. Operations and general anesthesia, however, are themselves factors known to have a strong effect upon the secretion of corticosteroids, and for this reason many changes in the function of the glands, possibly of importance in the pathogenesis of certain diseases, may remain unnoticed against the background of the operation, or their interpretation may be incorrect. The possibility of obtaining blood flowing from the adrenals in chronic experimental conditions is therefore of considerable interest, and has already attracted the attention of research workers. As long ago as in 1927, for instance, in experiments on dogs, P. I. Popov [1] introduced an angiostomy cannula by London's method into the vena cava at the point of entry of the lumboadrenal vein. When blood was taken through this cannula, however, the blood entering the syringe was mainly drawn from the vena cava, and it appeared impossible to obtain blood from the adrenals in this way.

The introduction of angiostomy cannulas of the most varied construction directly into the lumboadrenal vein was also unsuccessful on account of the small size of this vessel even in large dogs. A more reliable method was developed by the American workers Hume and Nelson [4] (Fig. 1a). The operation was carried out on a dog. The right lumboadrenal vein was ligated and divided above the adrenal gland. A polyethylene tube was tied into its central end and the tube brought out to the surface. Below the adrenal, at the mouth of the lumboadrenal vein, a clamp was applied in the form of a loop of polyethylene thread. Both ends of this thread were also brought out through the tube.

After suture of the wound, the loop was drawn tight enough to compress the lumboadrenal vein at its point of entry into the vena cava, so that the blood flowing from the adrenals through the cannula tied into the central end of the lumboadrenal vein flowed out to the surface. In the period between taking the blood, the cannula was closed with a cork and filled with heparin solution.

We were unable to obtain blood for more than 4-5 days after operation from animals treated in this manner.

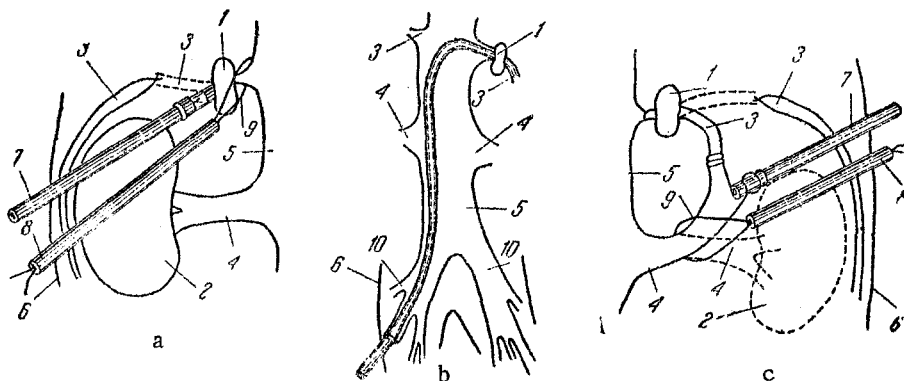


Fig. 1. Scheme of introduction of the cannula. a) By the method of Hume and Nelson; b) by the method of Endroczi et al.; c) after anastomosis of the renal and lumboadrenal veins; 1) adrenal gland; 2) kidney; 3) lumboadrenal vein; 4) renal vein; 5) inferior vena cava; 6) skin; 7) PVC cannula; 8) clamping tube; 9) clamp; 10) femoral vein.

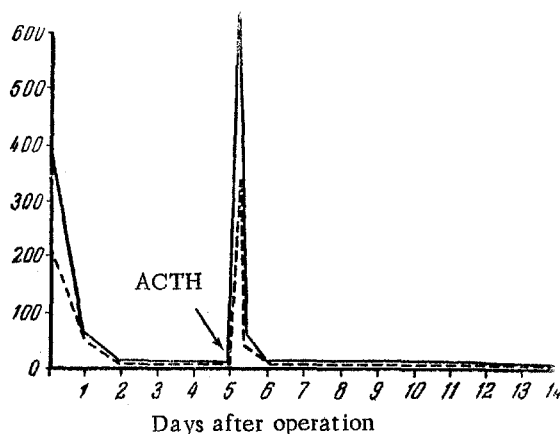


Fig. 2. Rate of secretion of hydrocortisone (continuous line) and corticosterone (broken line) in dogs after insertion of a cannula for the prolonged withdrawal of blood from the adrenal gland.

the external end of the catheter. In the intervals between taking the blood samples, the catheter was closed with a cork and filled with heparin solution. According to the authors of this method, the catheter may remain in the animal for 2-3 weeks. When blood is taken by this method, it is evidently possible for blood to be aspirated from the vena cava. If the blood is collected at a time of intensive secretory activity of the glands, this does not matter very much, for in calculating the rate of secretion it is important only that all the blood flowing from the adrenal gland during a known time should be obtained, and its dilution with peripheral blood is of no consequence. In those cases, however, when the gland is in a resting state and the secretion of corticosteroids is very slight, the dilution of adrenal venous blood with peripheral blood becomes undesirable. In our work, we have therefore, attempted to modify the method of Hume and Nelson in order to prolong the time during which adrenal blood may be obtained. Our suggested method is as follows (Fig. 1c).

The operation is carried out on large dogs (weighing 20-25 kg) on the left adrenal gland. After division of the abdominal muscles along the posterior edge of the last rib, the kidney is exposed. The ureter and renal arteries are ligated and divided. The renal vein is mobilized as far as its point of ramification. Ligatures are applied to both branches of the vein. The kidney is removed. The lumboadrenal vein is next dissected out. All veins entering it are ligated. The vein itself is ligated and divided above the adrenal. By means of an

Judging by the investigations known to us [2, 4-7], it has rarely been possible to obtain blood for a longer time. In our view a drawback of this method is that an "idle" portion of vein is formed between the cannula and the adrenal, and this rapidly becomes obliterated and atrophic.

Another method belongs to the Hungarian workers Endroczi, Bata and Martin [3]. Laparotomy was performed on a dog under general anesthesia, and a thin plastic, elastic catheter was then introduced through the femoral vein into the inferior vena cava (Fig. 1b). The catheter was passed along the inferior vena cava into the orifice of the lumboadrenal vein as far as the point where it received the adrenal veins. Fixation of the catheter in the required position was achieved by attaching it to the inner wall of the vena cava by means of a few fine silk sutures. The wound was closed. When the dog had recovered from the operation, the adrenal blood was drawn off as required through

apparatus for vascular suture the central end of the lumboadrenal vein is sutured to one of the branches of the renal vein. Into another branch is tied a polythene tube so that no "idle" areas of vein are left. The tube is brought out to the surface. The mouth of the lumboadrenal vein is ligated and to the mouth of the renal vein is applied a clamp of high-number "saturn" wire. By this method of introduction of the cannula, no "idle" areas of vein are formed. Throughout its whole extent, the walls of the vein are bathed with flowing blood. The operation is carried out at some distance from the adrenal, so that the gland remains free from trauma. The animals withstand the operation well. From dogs undergoing operation in this way, adrenal blood can be collected for 10-15 days after operation. In Fig. 2 the results showing the secretion of corticosteroids in one dog undergoing operation by this method are illustrated. It may be seen from Fig. 2 that the rate of secretion of both hormones - corticosterone and hydrocortisone - fell to a low level on the day after operation, and remained there throughout the period of investigation. The injection of 40 units of ACTH caused a sharp but transient rise in the secretion of both corticoids, with a maximum one hour after the injection of ACTH. It can be concluded from these findings that the adrenal function in dogs undergoing operation by the method described above rapidly returns to normal, and that the reaction of the glands to injection of ACTH is not affected.

#### SUMMARY

The author describes a method of a cannula implantation for procuring blood flowing out from the adrenal gland in chronic experiments on dogs. The method involves nephrectomy, uniting the lumboadrenal vein to one of the renal vein branches and introduction of polyethylene tube into another branch. This method permits blood to be obtained from the adrenal glands for a period of 10 to 15 days after operation.

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