PERFUSION OF ISOLATED DOG SKIN*

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Internal organs lend themselves easily to perfusion experiments. Important knowledge about intermediary metabolism of kidneys, liver, thyroid, intestines and adrenals has been obtained by application of this method.

It is rather difficult to apply the perfusion method to studies with skin because the skin is supplied by small arterial branches of the deep cutaneous arterial plexus, and there is no skin area sufficiently large which is supplied by one single artery. Malmejac and Desanti (1) in 1938 called attention to the fact that the skin of the inner aspect of the upper hind limb of the dog is readily detached and receives its arterial suppply from a distinct isolated saphenous artery. In 50 dissections on dogs Malmejac et al. found this artery always to be present. They were unable to find an analogous artery in man. This anatomical finding made it possible to study cutaneous circulation in this area of the dog skin by cannulating the saphenous artery and saphenous vein (2), (3), (4), (5).

In this report the observations of these workers are confirmed (Fig. 1) and a new technic for perfusing an isolated piece of dog skin in an incubator box is described.

EXPERIMENTAL

A medium size lean dog with little subcutaneous fat is selected. Intravenous nembutal is given. The dose of nembutal is approximately 60 mg. (1 gr.) per 5 lbs. The medial aspect of the upper hind limb is shaved. An incision through the skin parallel to and approximately 2 cm. distal to the groin is made. The incision runs from the anterior to the posterior border of the medial aspect of the leg. The site of this and subsequent incisions is shown in Figure 2.

Slight oozing from skin edges is controlled with electrocoagulation or ligatures.

The next incision is made following the anterior border of the thigh from the upper incision to 2 cm. below the knee. The third incision follows the posterior border from the upper incision to 2 cm. below the knee. The upper half of the cut skin is detached from the subcutaneous tissue by lifting it with a slight pull. Now the saphenous artery, vein and nerve are easily seen embedded in the subcutaneous tissue of the leg. The branches given off from the lower parts of artery and vein supply the detached skin area.

Next the saphenous vessels are dissected free, ligated and cut across distally to the ligature midway between groin and knee. Occasional small muscular

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Read before the Fourteenth Annual Meeting of The Society for Investigative Dermatology, Inc., New York, New York, May 30, 1953. (With presentation of a 16 mm. colored moving picture.)

This work was supported, in part, by a Research Grant from The Toni Company, Chicago, Illinois.

branches are ligated. The rest of the partly detached skin is now further separated so that the skin flap is connected to the leg with only a narrow stalk 2 cm. below the knee. Because the saphenous vessels were dissected free from the subcutaneous tissue the vessels slip off with the skin flap when it is detached as its branches to the skin within this area. More distally the saphenous vessels supply skin and muscles of the lower leg. The vessels are again ligated 2 cm. below the knee. Finally the skin flap is cut off at this point.

Immediately after the skin piece is removed it is fixed with the help of clamps and rubber bands on a supporting frame and placed in an incubator box under constant temperature and humidity for the perfusion experiments (Fig. 3).

Canulae (hypodermic needles, No. 18 or 20) are then inserted into the upper parts of the saphenous vessels (Fig. 4) and perfusion is started immediately. Because the lower end of the saphenous vessels have been ligated the blood will

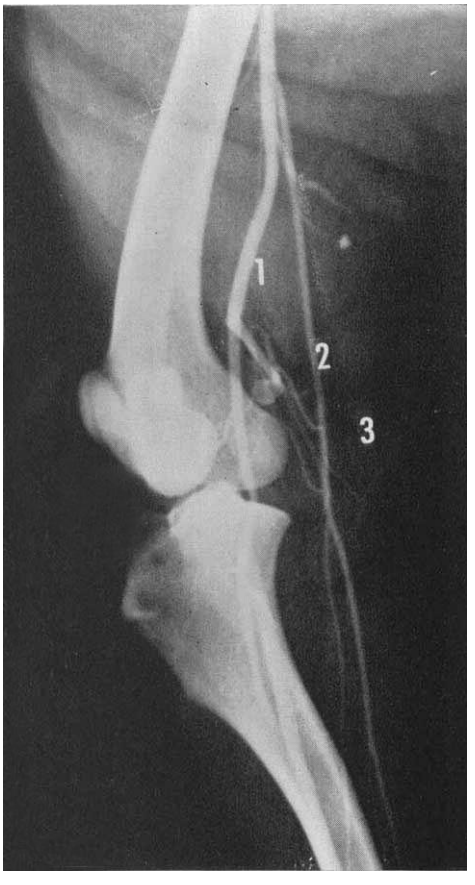

FIG. 1. X-ray picture of femoral and saphenous vessels after injection of radio opaque material. 1. Femoral artery. 2. Saphenous artery. 3. Cutaneous branches of saphenous artery.

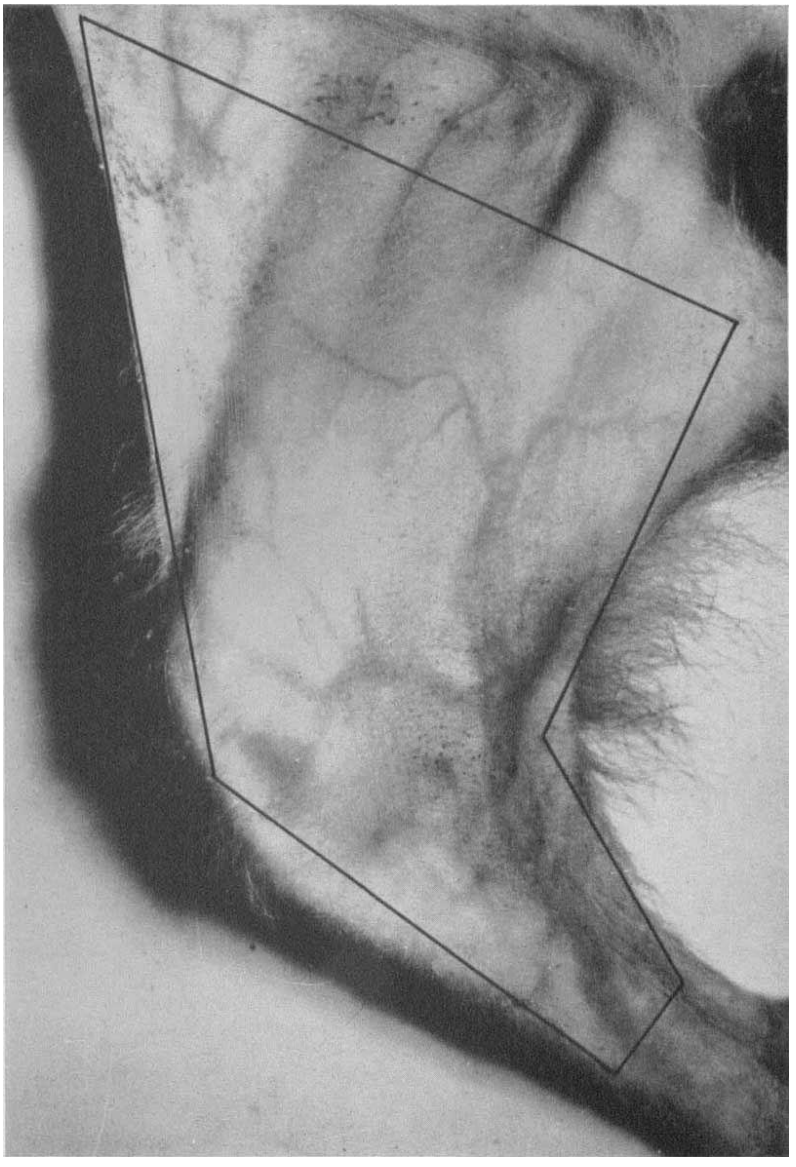

FIG. 2. Infra-red photograph of medial aspect of thigh showing a portion of the saphenous vein system. The skin area described in the text is outlined.

be forced to circulate through the cutaneous branches only and return through the accompanying venous system of the same area (Fig. 5).

The removed skin piece of a medium size dog has approximately the size of a man's palm.

In these experiments the perfusion was carried out with heparinized dog blood

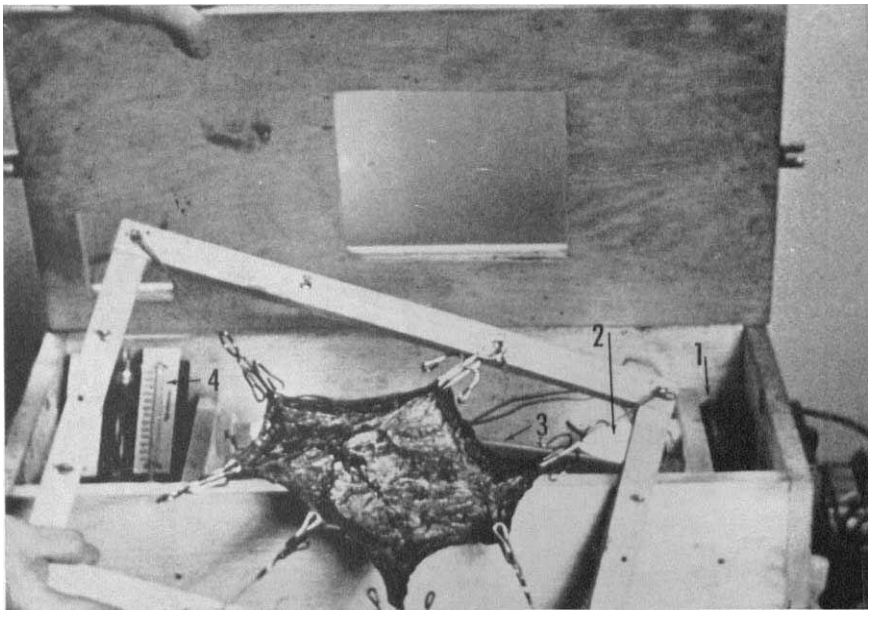

FIG. 3. Incubator box with lid open. The supporting frame holding the skin piece is ready to be placed into the box. 1. Thermoregulator. 2. Heater-bulb. 3. Water pan. 4. Thermometer and hygrometer.

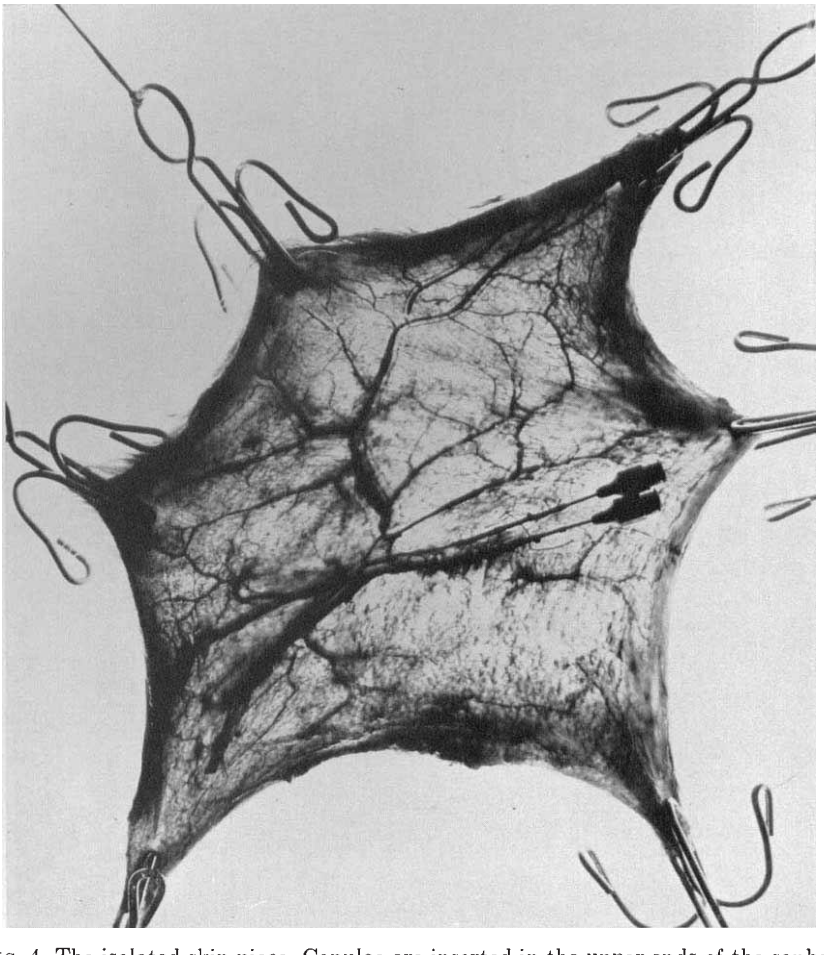

FIG. 4. The isolated skin piece. Canulae are inserted in the upper ends of the saphenous vessels. Lower ends of saphenous vessels are ligated.

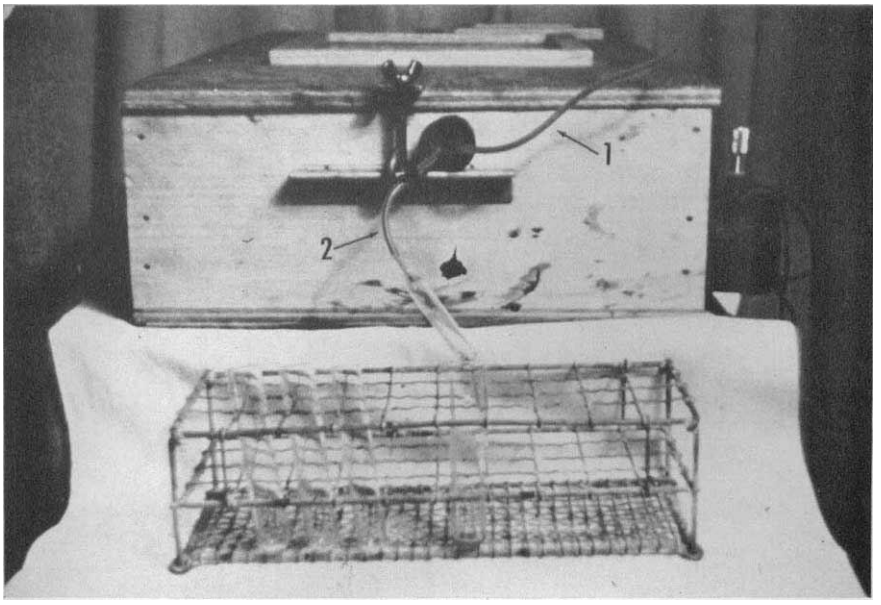

FIG. 5. Incubator box with lid closed. 1. Tubing for perfusion apparatus to the saphenous artery. 2. Tubing from the saphenous vein for collection of blood samples.

by a simple blood transfusion apparatus (Fig. 6). It is easy to regulate pressure at the arterial and venous end of such a gravity-operated apparatus by raising or lowering the respective blood fluid levels.

DISCUSSION

The operative procedure is simple. Removal of the skin piece takes only 15 to 20 minutes. Perfusion experiments with a steady pressure have been run with heparinized dog blood circulating for more than three hours.

The importance of a well balanced systolic-diastolic pressure system in perfusion experiments on surviving organs is discussed in detail by Pappenheimer (6). Different types of "artificial hearts" have been described in the literature to simulate normal hemodynamics in perfusion experiments (7). A blood pump for whole blood without anticoagulants was described recently by Osborn (8). Work is in progress in our laboratory to adapt Osborn's method for the perfusion of the isolated piece of skin.

The described skin preparation lends itself to the study of different aspects of skin metabolism: 1. The fate of material added to the arterial blood can be studied by analyzing the outflowing venous blood. 2. Material added to the arterial blood may be carried to the skin surface by diffusion, secretion or excretion through glandular or epidermal activity and may be recovered there unchanged or in form of metabolites by wiping the surface. 3. Percutaneous absorption can be studied by applying material to the surface and analyzing the outflowing venous blood. Such studies can most easily be carried out with the use of radioactive tracers.

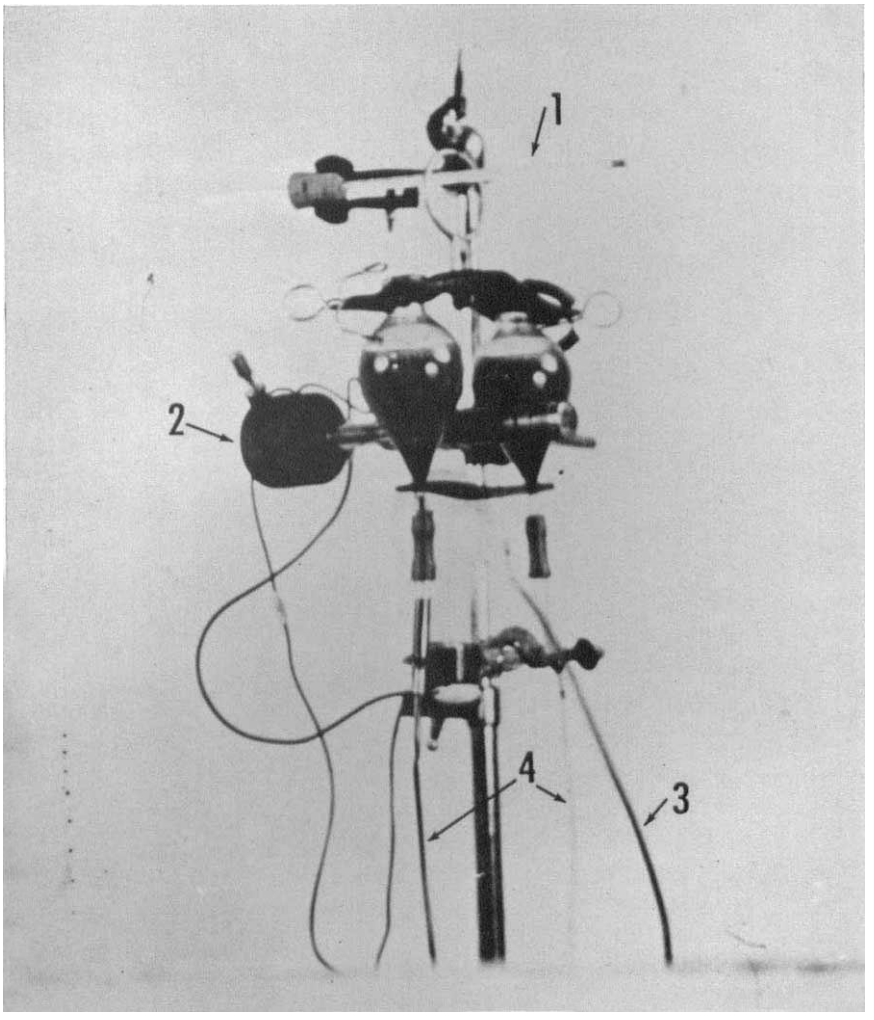

FIG. 6. Picture showing perfusion apparatus. 1. Thermometer. 2. Thermoregulator. 3. Tubing from bottles to saphenous artery. 4. Cord connected to shaker to keep the blood agitated.

SUMMARY

A technic for perfusion of isolated dog skin is described. Perfusions were carried out for more than three hours with this preparation. Its application for metabolic studies is discussed.

Addendum: After this manuscript went to print, I found that Feldberg and Paton used a somewhat similar experimental arrangement in the cat. (Feldberg, W. and Paton, W. D. M.: Release of histamine from skin and muscle in the cat by opium alkaloids and other histamine liberators. J. Physiol., **114:** 490–509, 1951.)

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