STUDIES ON ORGAN TRANSPLANTATION.

II. TRANSPLANTATION OF THE SPLEEN WITH INTACT BLOOD SUPPLY.

BY K. KAWAMURA, M.D.

(From the Division of Experimental Surgery of the Mayo Clinic, Rochester, Minn.)

(Received for publication, April 4, 1919.)

During the last 15 years organ transplantation has been studied extensively with especial reference to practical therapeutics and the biologic possibility of regeneration of transplanted tissues.

Many experiments have been reported on the ductless glands, and it has been affirmed that in these experiments the healing of the grafts depends largely on the site of the transplantation, and on the size of the transplant. As to the location of the graft, the opinions of authors differ widely. Cristiani, Ribbert, and others believe that the subcutaneous connective tissue is the best soil for the transplantation of the thyroid tissue; Payr preferred the spleen, while Kocher used the bone marrow. With regard to the size of the transplanting piece Cristiani, Ribbert, and Lubarsch claimed that numerous small fragments of thyroid tissue, the size of a wheat grain, are preferable. Payr, Kocher, von Bramann, and others advised the use of large grafts the size of a hazelnut or cherry because the larger portions undergo destruction.

Many instances of transplantation of the thyroid, parathyroid, ovary, and adrenals are found in the literature, and in some the results were favorable, although the percentage of success was small. On the other hand, reports of the transplantation of the spleen are rare, and the results have been unsatisfactory compared with those of the other ductless glands.

In 1916 Manley and Marine published an article on the transplantation of ductless glands. They transplanted six autografts and eighteen homografts of splenic tissue in the subcutaneous tissue of the abdomen, all of which were absorbed in 12 days. In 1917 they published another article on spleen transplantation. This time twelve attempts at homotransplantation and six attempts at autotransplantation were made on fifteen rabbits. The method employed was the same that was formerly used by them, and consisted of transferring small sections of the spleen of about 2 mm. into the subcutaneous fascia of the abdomen. None of their attempts at homotransplantation was successful beyond the usual results persisting for 2 or 3 weeks, common to all homografts. They ob-
tained only one successful permanent autotransplantation. The graft was removed at autopsy 325 days after transplantation and showed all the morphologic characteristics of a fully differentiated and functionally active spleen. The writers stated that considerable difficulty in spleen transplantation may be due to its complex anatomic structure.

The failure of transplantation of pieces of the organ, however, is partly attributable to insufficient blood supply in consequence of which the grafts are absorbed by degrees. Besides, it should be taken into consideration that small pieces, although successfully transplanted, are not always sufficient for the deficient functions. The transplantation of the entire organ by anastomosis of its blood vessels to suitable parts of the circulatory system can yield sufficient nutrition and probably also function.

Lüdke tried to transplant the spleen of dogs into the splenic vessels of a goat and a wether by blood vessel anastomosis. In all three instances there was faulty blood vessel anastomosis. He attributed the failure in the use of the technique to the unfavorable location of the spleen of these animals, and to the fact that the lumen of splenic vessels is too small. Lüdke, therefore, changed his first plan and transplanted pieces of spleen of rabbits or dogs into the abdominal cavity, the stomach wall, or into the spleen of rabbits, dogs, monkeys, goats, or wethers. The transplanted splenic tissue was observed for 4 weeks, but after 2 or 3 months no part of it was visible.

Carrel extirpated the spleen, washed it out with Locke's solution, and replaced it by blood vessel anastomosis in two dogs. One spleen necrosed and completely disappeared, due to the occlusion of the artery. The replantation of another spleen was successful, so that the anatomic condition of the vessels and of the organs was entirely normal about 8 months after the operation.

EXPERIMENTAL.

The entire series of our experiments was performed on dogs. There are usually two trunks of both artery and vein to the spleen from the gastrosplenic vessels. One pair enters the spleen in its lesser (left) end and another almost in the middle. The spleen was divided in two parts, corresponding to the stream district of these large branches, after the mattress sutures had been applied transversely on it. The half of the spleen which is nourished by the larger branches was used for the transplantation. The attached omentum was cut off after ligation. The splenic artery, vein, and nerves were dissected and
divided, Crile clamps being used. The caliber of the artery was hardly 1.5 mm. in diameter. The spleen was then removed, and wrapped up in a salt sponge. After a few minutes the spleen was replaced into the abdominal cavity, and its vessels were united as before, by end to end anastomosis. In one case an attempt was made to transplant the spleen into the neck. After the thyroid had been removed, the peripheral end of the splenic artery was united to the central end of the superior thyroid artery and the peripheral end of the splenic vein to the central end of the external jugular vein. In another case the spleen was transplanted into the renal vessels after nephrectomy. Most of the experiments were performed autoplastically, but in one instance the spleen from one animal was transplanted to another. The arterial suture was always difficult, due to the small caliber of the vessel. As soon as the clamps were unfastened, the bluish red collapsed spleen became very red, and its volume was increased. The circulation in the spleen was reestablished between 1 and 2 hours after its extirpation. The dissected omentum was reunited, and in a few cases the nerves also were sutured. Several days after the operation the condition of the transplanted spleen was ascertained by laparotomy. When the transplantation was successful, the other intact half of the spleen was removed and immersed in a jar filled with 10 per cent formaldehyde solution for microscopic examination, and the animal was observed further.

The protocol of a successful experiment is given below.

Experiment 1 (Dog 1).—Adult, mongrel bulldog, male; weight 16.9 kilos.
Oct. 5, 1918. Operated on under intratracheal ether narcosis. The spleen was exposed through a left rectus incision. Two rows of mattress sutures were placed in the middle of the spleen and the organ was divided between them. One half of the spleen, which was nourished with smaller branches of the splenic artery, was left in the abdominal cavity; the other half was used for replantation. After the dissection of the nerves and omentum, the artery and veins were cut so that this half of the spleen could be brought out extraperitoneally, apart from its connection. The spleen was wrapped up in a salt sponge for a few minutes and replanted into its original site by end to end blood vessel sutures. The arterial lumen measured hardly 1.5 mm. in diameter, and suturing was difficult. The venous suture, on the contrary, was easy. The clamp on the vein was first unfastened and then the clamp on the artery. The spleen, which during the operation appeared dark blue, and was contracted, became normal in color, and as-
<table>
<thead>
<tr>
<th>Experiment No.</th>
<th>Operation,</th>
<th>Duration of observation,</th>
<th>Results,</th>
<th>Remarks,</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>days</td>
<td>Condition of blood vessels,</td>
<td>Condition of transplanted spleen,</td>
</tr>
<tr>
<td>2 (&quot; 2).</td>
<td>Oct. 7. Spleen which was divided in a proportion of one-third and two-thirds by mattress suture re-planted, combined with nerve suture.</td>
<td>3</td>
<td>Thrombosis in central part of arterial suture. A small part at venous suture leaked.</td>
<td>Necrotic; softened.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>---</td>
<td>---</td>
<td>---</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5 (Dog 5).</td>
<td>Oct. 16. After removal of left thyroid gland, half of spleen transplanted to neck. Splanic artery united to thyroid artery and splanic vein to external jugular.</td>
<td>7</td>
<td>Both transplanted artery and vein thrombosed.</td>
<td>Necrotic; softened.</td>
</tr>
<tr>
<td>6 (&quot; 6).</td>
<td>Oct. 23. Replantation of half of spleen. Nov. 27. At exploratory laparotomy a large cystic tumor was found. Jan. 24, 1919. Extirpation of tumor.</td>
<td>93</td>
<td>Obstruction of artery at united part.</td>
<td>A cystic tumor about size of a child's head grew from replaced spleen. The cyst was filled with a muddy, purulent fluid.</td>
</tr>
</tbody>
</table>
TABLE I—Concluded.

<table>
<thead>
<tr>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>8 (Dog 8).</td>
<td></td>
<td>1918</td>
<td>61 days</td>
<td>Obstruction of artery at united part.</td>
<td>At second operation it was proved that the transplanted spleen kept its original size, but by incision no fresh blood flowed out. At third operation no part of spleen was visible.</td>
</tr>
</tbody>
</table>
sumed its original size as soon as the clamps were removed. The circulation through the replanted spleen was reestablished after an interruption of 1 hour. No suture of the omentum or the nerves was made. After the circulation through the transplanted vessels as well as the spleen was assured, the abdominal wound was closed. The first incision was made at 9.14 a.m.; the last stitch was inserted at 11.20 a.m.

Nov. 27 (53 days after the first operation). An exploratory laparotomy was done. The weight of the animal was 17.7 kilos.; there were no marked adhesions; the omentum was normal. The replaced spleen had kept its original size and appeared entirely normal in color and consistency, but its capsule was thickened. The other intact half of the spleen was extirpated.

Dec. 27. The animal fought with another dog and was seriously wounded in the abdominal wall, back, and both hip regions.

Jan. 1, 1919 (88 days after the first operation). The dog died of his wounds.

Autopsy.—The wounds that had been made by fighting were found to be infected; there was no infection of the abdominal cavity. The replaced spleen looked very well. No thrombosis could be found in the splenic artery or vein; their lumina were patent throughout. The suture lines on the vessels could be found with difficulty. They were covered with normal epithelium and were smooth.

Microscopic Examination.—The capsule was quite thick in places as compared with the control specimen. Otherwise the structure of the pulp, the number and the size of the trabeculae, and the Malpighian bodies were perfectly normal (Figs. 1 to 4).

Seven attempts at autoplastic transplantation and one attempt at homoplastic transplantation of the spleen were made, as shown briefly in Table I. Five of the seven autoplastic transplants were replanted to the splenic vessels, one to the neck, and one to the renal vessels. In two of the five replanted cases the dogs died in 3 and 4 days after the operation. At autopsy it was found that a great deal of bloody serous fluid filled the abdominal cavity, and that the spleen was necrosed, due to thrombosis. Two cases were examined 30 and 35 days after the operation by exploratory laparotomy. One half of the spleen was found intact in each case, but the other replaced half could not be seen. In one of these two instances there was about 1 liter of pale red, muddy, serous fluid in the abdominal cavity. A round, partly soft, partly hard cystic lump about the size of a child's head lay in the center of the cavity. This lump was surrounded by vast distended blood vessels and muddy gelatinous membrane. Later it was proved at autopsy that the lump was a cyst which had been formed by replanted spleen. The result of one operation (Exper-
ment 1), in which the spleen was replanted in a dog, was highly satisfactory. 88 days after the operation the spleen, as well as the united blood vessels, was in good condition. The spleen which had been transplanted into the neck was examined 7 days after the operation, and it was found that the whole tissue had undergone necrosis. The transplantation of the spleen into the renal vessels was troublesome, because the field of the operation was too deep and the caliber of the splenic artery was very small. Nevertheless, the immediate result of the operation was successful. As soon as the clamps were removed, the spleen became red and greatly distended. 14 days after the operation, when the exploratory laparotomy was made, the spleen retained its original size; but the color was pale red, and when it was incised dark blood came out. 47 days later the spleen had completely disappeared. The spleen which was grafted homoplastically was examined at autopsy 2 days after the operation. It was soft, dark red, and necrotic. The transplanted splenic vein was thrombosed. Hemorrhagic serous fluid filled the abdominal cavity.

DISCUSSION.

The results of the experiments immediately after operation were in all cases satisfactory. In spite of the interruption of the circulation from 1 to 2 hours after extirpation, the immediate circulation of blood through the transplanted spleen and blood vessels was favorable, but most of the spleen became necrotic or was entirely absorbed. The cause of this was the obstruction in the transplanted vessels, due to thrombosis. It is probably difficult to obtain good results by using so small a vessel as a branch of the splenic artery. Carrel has noted that such a small vessel cannot be sutured with many chances of success. As has been stated, all Lüdke's experiments failed.

Regardless of these difficulties, our successful case showed that such a highly differentiated, complicated organ as the spleen can be transplanted en masse, that it can keep permanently its normal structure, and probably also can functionate normally. In this instance the difference between the central and peripheral parts of the grafts, as Manley and Marine experienced by piecemeal transplantation, was not visible. This is scarcely to be expected because in trans-
planting by blood vessel sutures the nourishment of the transplant is maintained throughout.

In view of the fact that the spleen can survive even if the nerves are not united, the experiment demonstrated that nerves are not essential for the maintenance of grafts.

The neck, probably also the inguinal furrow, is not a favorable site for the experimental transplantation of the spleen by blood vessel suture, because after the suture of the fascia and skin the more or less distended spleen is compressed, and, consequently, disturbances of the circulation through the graft may occur.

SUMMARY.

Seven autoplastic transplantations and one homoplastic transplantation of the spleen of dogs were made. One autotransplantation was successful, the gland being normal at the end of 88 days.

BIBLIOGRAPHY.

Cristiani, quoted by Lüdke.
Kocher, T., Ueber die Bedingungen erfolgreicher Schilddrüsentransplantation beim Menschen, Arch. klin. Chir., 1914, cv, 832.
Lubarsch, quoted by Lüdke.
Ribbert, quoted by Lüdke.
EXPLANATION OF PLATES.

PLATE 7.

Fig. 1. Photomicrograph of the intact portion of the spleen of Dog 1. × 50.
Fig. 2. Photomicrograph of the same specimen as Fig. 1. × 125.

PLATE 8.

Fig. 3. Photomicrograph of the transplanted portion of the spleen of Dog 1. × 50.
Fig. 4. Photomicrograph of the same specimen as Fig. 3. × 125.
Fig. 1.

Fig. 2.

(Kawamura: Organ transplantation, II.)
Fig. 3.

Fig. 4.

(Kawamura: Organ transplantation. II.)