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Hemodynamic Effects of Renal Transplants in Hypertensive and Control Rats.* (28813)

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From several types of studies it appears that transplantation of a normal kidney reduces arterial pressure in a hypersentitive recipient. In dogs made hypertensive by bilateral nephrectomy and a diet rich in protein and salt, kidney transplants were able to decrease arterial pressure in less than 24 hours, without a decrease in the fluid compartment(1). In 3 patients with renal hypertensive disease, kidney homotransplants from normal twins produced a decrease in arterial pressure. This occurred even when the diseased kidneys were left in place(2). In the rat with arterial hypertension developed by DCA treatment and in the renal hypertensive rat, transplantation of a normal kidney lowered the arterial pressure to normal levels within about a half hour(3).

This report concerns the hemodynamic change responsible for the decrease in arterial pressure produced by a normal kidney transplant in a hypertensive rat. Experiments were done in which we measured the changes in arterial pressure, cardiac output, and resistance in a vascular bed in normotensive and renal hypertensive rats before and after transplantation of a normal kidney.

Methods. Twenty-five male Sprague-Dawley rats weighing between 250 and 410 g were used. Fourteen were made hypertensive by placing a figure-of-8 ligature on one kidney and one week later removing the remaining kidney. In the control animals only the unilateral nephrectomy was performed. Arterial pressure was measured at intervals using the tail microphone method(4). Four weeks following application of the ligature the animals were hypertensive.

From 7 to 9 weeks after the figure-of-8 ligature was placed on the kidney, changes in hind limb resistance and cardiac output caused by transplantation of a normal kidney were studied. Details of the methods are summarized in the legends of Fig. 1 and 2. In 6 hypertensive and 6 control rats the resistance in the hind limb perfused with autogenous blood at a constant rate of 1.5 ml/min was observed. In 8 hypertensive and 5 control animals cardiac output was measured with the thermodilution indicator method. The animals were anesthetized with sodium pentobarbital (40 mg/kg, i.p.), tracheotomized heparinized (10 mg/kg, i.v.). Systemic arterial pressure during the experiment was monitored by a centrally directed cannula in the femoral artery. All the parameters were recorded on a Grass polygraph.

The technique for kidney transplant has been described(3). Coagulation of the blood in the renal vascular tree was avoided by replacing blood with a heparin solution in saline. The ischemic period of the kidney never exceeded 20 minutes. The transplantation procedure caused an immediate random variation in arterial pressure; hence, the 5- to 10-minute period required for this procedure was excluded from the data presented here.

Experimental observations were made as follows: after control period, A, lasting 10-30 minutes, the normal kidney from a donor rat was transplanted to the recipient's femoral

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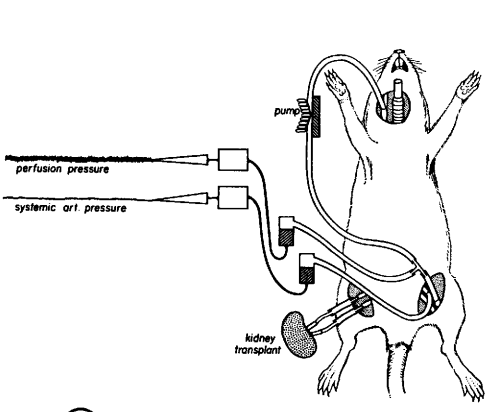

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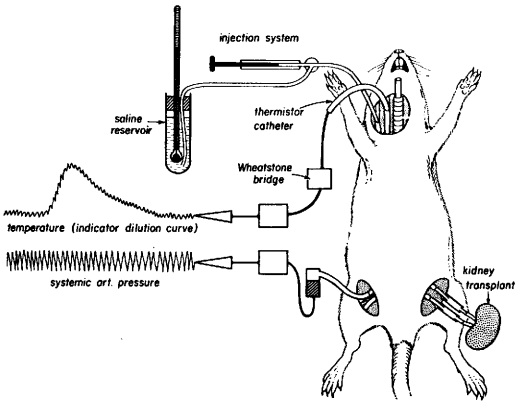

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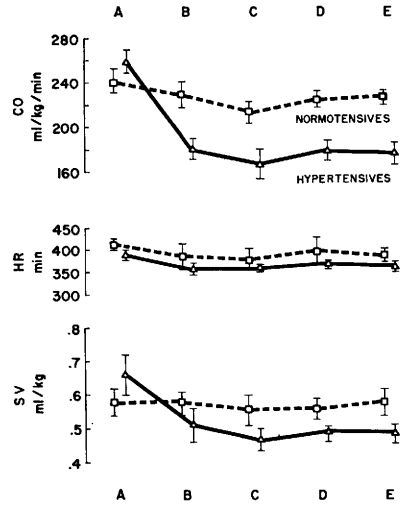

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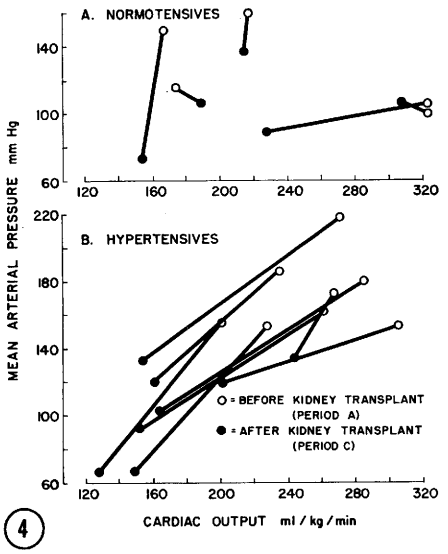

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FIG. 1. Kidney transplant to rat with one limb perfused with autogenous blood. Blood withdrawn from a carotid artery is pumped at a constant rate into femoral artery of perfused limb. A T-tube near the perfused limb joins the system to a transducer from which perfusion pressure is recorded via a Grass polygraph. A centrally-directed cannula in the same femoral artery is joined to a second transducer to record systemic arterial pressure. Kidney is transplanted to femoral artery and vein of the contralateral leg.

FIG. 2. Thermodilution technique for measurement of cardiac output. A closed system for injection of indicator is at room temperature (20-25°C). Saline (0.1 ml) is withdrawn from the reservoir, where its temperature is known to the nearest 0.1°C, into a Hamilton microliter syringe; it is then injected rapidly into the rat. Dilution of the indicator (change in temperature) is recorded in the ascending aorta by a thermistor (Fenwall Electronics, GC 32 J1) which has a time constant in saline of 0.17 sec. Temperature and arterial pressure are recorded on a Grass polygraph. The kidney transplant is made to femoral artery and vein of one of the legs.

FIG. 3. Effect of kidney transplant on cardiac output (CO), heart rate (HR), and stroke volume (SV). Letters on horizontal axis represent the various periods of experiment. Values are averages for each time period: A = 15-0 min prior to kidney transplant (a subsequent 5-10 min period required for manipulation of transplant is omitted); B = 0-15 min after completion of transplantation; C = 16-30 min after completion of transplantation; D = 0-15 min after transplant removal; E = 16-30 min after transplant removal. Vertical bars represent SEM. Kidney transplant produced precipitous falls in CO and SV in hypertensive rat.

FIG. 4. Role of cardiac output in blood pressure change produced by kidney transplant. A illustrates the random relation between changes in cardiac output and mean arterial pressure in normotensive rat following kidney transplant. Kidney transplant in hypertensive rat (B) elicited uniform and parallel decreases in both these parameters indicating that in this group the fall in CO caused the decreases in arterial pressure.

artery and vein and 5-10 minutes were allowed to elapse for stabilization of the preparation. Data from the next 15-minute period, B, and the following 15-minute period, C, were averaged individually. At the end of period C the circulation between the rat and the kidney transplant was interrupted by a clamp and 2 additional 15-minute intervals were studied, D and E. Since periods B and D were transition periods, attention was focused mainly on the differences obtained between periods A and C, and between C and E. Hemodynamic parameters were measured arbitrarily at 5-minute intervals during each period. Therefore in each rat the values quoted represent a mean of 3 or 4 readings.

Results. Action of kidney transplant on mean arterial pressure (MAP) of control and hypertensive rats. The average MAP in the 14 hypertensive rats was 170.6 ± 5.0 (S.E.M.) mmHg and in the 11 control rats, 138.1 ± 7.0. The kidney transplant in the hypertensive animals produced a marked decrease in MAP of —57.2 ± 10.0 mmHg between periods A and C. This decrease was highly significant ($p<.001$). In control rats MAP also decreased, —24.6 ± 8.0 mmHg between periods A and C. This decrease was also significant ($p = .008$). The decrease in MAP produced by transplantation of the kidney was significantly greater in the hypertensive than in the normotensive rats ($p = .01$).

After the kidney transplant was eliminated, the average MAP increased in both groups, but the change was not statistically significant.

Pressure in the hind limb perfused with autogenous blood. Perfusion pressure, and therefore vascular resistance, in the hypertensive rat was not different from that of the control, 121.6 ± 11.3 mmHg and 122.7 ± 11.1, respectively. Perfusion pressure tended to increase in both control and hypertensive animals throughout the experiment: +16.5 ± 12.1 mmHg from period A to C in the hypertensive rats and +21.3 ± 11.1 in the controls. Neither increase seemed to be caused by the kidney transplant nor was it statistically significant. During the 30 minutes following removal of the transplant the vascular resistance in the leg did not return to control level.

Action of kidney transplant on cardiac output (CO) and total peripheral resistance (TPR). The average CO in the 8 hypertensive rats was 258.0 ± 10.4 ml/kg/min and in the 5 controls 240.5 ± 10.7. Between periods A and C the kidney transplant produced a great decrease in CO in the hypertensive animals (—91.6 ± 11.9 ml/kg/min, $p < .001$). This corresponded with a parallel change in MAP. In the normotensive rats, the kidney transplant produced a lesser decrease in CO between periods A and C: —25.7 ± 20.5, but it was not statistically significant ($p = .30$). The data are presented graphically in Fig. 3 and 4. The reduction in CO in hypertensive rats was significantly greater than the decrease produced in control animals ($p = .02$). The value for CO in the hypertensive animals fell to 165.3 ± 13.8, or well below the value obtained by this method in normal rats. After the kidney transplant was removed, a slight recovery in CO was seen in both groups of animals, but it was not statistically significant.

Calculated TPR in the hypertensive rat, .682 ± .030 PRU/kg, was higher than in the normotensive rats, .603 ± .111, but the difference was not statistically significant. The transplant produced a slight but not significant reduction of TPR in both groups between period A and C, —.045 ± .139 PRU/kg in the hypertensives and —.124 ± .90 in the controls. As in the measurements of regional resistance in the hind leg, changes in calculated total resistance were negligible.

Action of kidney transplant on heart rate (HR) and stroke volume (SV). Reduction in HR (Fig. 4) was on the average equal in the control and hypertensive groups, but only in hypertensive animals did it reach significant levels ($p > .018$ in hypertensive rats between periods A and C and $p = .230$ in the controls). SV fell greatly in the hypertensive rats (Fig. 4), —.195 ± .040 ml/kg ($p = .004$), while no changes in SV occurred in the control animals after transplantation, —.023 ± .05 ml/kg ($p = .673$). The difference in the effect of kidney transplant on SV, between hypertensive and control ani-

mals, was significant ($p = .025$).

Action of kidney transplant on pulse pressure (PP). PP in the hypertensive rat was 37.1 ± 5.15 mmHg and in the control 34.4 ± 1.2. The kidney transplant produced a large decrease in PP in the hypertensive animal between period A and C, −14.1 ± 4.6 mmHg, which is statistically significant ($p = .024$), and a small decrease in control animals in the same interval, −1.4 ± 2.8. After the transplant was eliminated some recovery in PP was seen in the hypertensive group, but it was not statistically significant.

Discussion. From this study it is clear that in a hypertensive rat the decrease in MAP produced by transplantation of a normal kidney is due to a decrease in CO. If there is a decrease or increase in TPR it is not large enough to be an important influence on the change in MAP. The slight but significant decrease in MAP seen in control rats was caused by a decrease in both CO and TPR.

We have no explanation for our failure to observe a significant recovery in MAP and CO during the 30 minutes following exclusion of the kidney transplant. In other studies(3) a rise in MAP after exclusion of the kidney has been observed.

There was a slight decrease in HR and a large decrease in SV when the kidney was transplanted into a hypertensive rat; this was much less evident in the normotensive recipient. Although these experiments do not clarify the mechanism that produces these changes, Hajdu and Leonard(5) have described a cardioglobulin system in the blood which has a positive inotropic effect on the heart and which is increased in human hypertension. If such a system is also operative in the hypertensive rat, it is possible that the normal kidney transplant reduces these cardioglobulins to normal levels. Other possibilities are that the kidney transplant produces a greater inhibition of the sympathetic nervous system in the hypertensive rats or that it produces a substance which has a negative inotropic effect on the myocardium of the hypertensive animals.

The substance of renal origin responsible for reduction of MAP and CO in a hypertensive rat may be similar to the material extracted from the medullary portion of the kidney(6) which has been shown to decrease arterial pressure in hypertensive animals but not in normals. This principle has recently been greatly purified(7).

Our findings are similar to those of Ledingham and Cohen(8) that in the hypertensive rat with clipped kidney, when the renal arterial clip is released and the kidney perfusion improves, a decrease in arterial pressure occurs which is also mediated by a fall in CO. They proposed that these changes may reflect the reversal of the mechanism which brings about the development of hypertension.

Since completing our study we have learned of a similar study by Blaquier and Taquini (9). In addition to studying the effects of kidney transplant on renal hypertensive and control rats, these authors also utilized DCA-hypertensive rats. Resistance was studied in the perfused hind limb, and cardiac output was measured by the RISA indicator dilution technique. These authors observed no difference in the response of DCA- and renal-hypertensive animals to the kidney transplant. Since our results are in accord with those of Blaquier and Taquini, the conclusions of both are fortified.

Our own studies, those of Ledingham and Cohen, and those of Blaquier and Taquini introduce the possibility that in addition to the renin angiotensin system which affects chiefly TPR, there is also a renal mechanism concerned with the control of CO. This mechanism may also play a role in the pathogenesis of renal hypertension.

Summary. Transplantation of a normal kidney to a renal hypertensive rat produced a decrease in mean arterial pressure of 171 to 138 mmHg and a decrease in cardiac output from 258.9 to 167.3 ml/kg/min. There was no significant change in calculated total peripheral resistance or in regional vascular resistance in the perfused hind leg. The decrease in mean arterial pressure is caused by a decrease in cardiac output resulting from a decrease in stroke volume. Kidney transplantation to a normal rat elicits a lesser decrease in mean arterial pressure, which is caused by a decrease in both cardiac output and in total peripheral resistance.

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Induction of Tolerance to Skin Grafts in Mice with Disrupted Liver and Kidney Cells.* (28814)

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A specific state of tolerance to homografts across both weak and strong histocompatibility barriers may be induced in adult inbred mice by a variety of techniques including parabiosis and injection of large quantities of cells of donor origin(1-13). More recently it has been shown that cell-free antigenic material prepared by cell disruption is similarly capable of inducing tolerance in adult mice (14-18). This finding may be considered as further supporting the views previously expressed from this laboratory that fundamentally all experimental techniques for induction of tolerance are similar and that the main factors are antigenic disparity between host and donor, the relationship between quantities of donor antigen and mass of immunologically competent cells in the host, and rate and duration with which antigen is presented to the host. Agents such as X-radiation or drugs that suppress immune response may be viewed as means whereby the mass of lymphoreticular tissue is reduced or is prevented from proliferating in response to antigen. Under these circumstances relatively small quantities of antigen may be sufficient to overwhelm or exhaust the immunologically competent cells of the host with the resultant induction of tolerance. Contrariwise, the same end may be achieved if enormous doses of antigen can be injected into the host so that the lymphoreticular cell mass is overwhelmed in spite of its ability to proliferate in response to antigen. The latter technique would have much to recommend it clinically as well as experimentally since it would avoid the generalized and non-specific destructive effects of irradiation or suppressive drugs on the host including such undesirable complication as increased susceptibility to infection, impairment of general nutrition and damage to reproductive and other tissues. Even more important, such an approach using disrupted cellular material would obviate the occurrence of a graft-*vs*-host reaction attributable to viable immunologically competent cells with its attendant morbidity and mortality. The technique of employing disrupted cell materials as the tolerance-inducing antigenic source is admittedly more difficult than the use of intact replicating cells since larger amounts of material must be used and this material must be presented to the recipient over longer periods. In addition, the necessary quantity of antigen may be essentially unattainable if one has to rely on the use of spleen or bone marrow cells in non-inbred lines of animals since here all the antigen must be obtained from one donor. It seems likely that the antigens involved in histocompatibility re-

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