The Prevention of Acute Tubular Necrosis in Renal Transplantation by Chronic Salt Loading of the Recipient[1]

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By replacing normal water intake with 0·9% saline solution on a continuing basis, animals are protected against acute ischæmic tubular necrosis. Last-minute plasma volume expansion does not provide this protection. To see whether the protection is intrinsic to the kidney or merely involves the host, kidneys were transplanted syngeneically from salt-drinking rats to water-drinking rats, and vice versa, and the recipients were injected with glycerol to produce myohæmoglobinuric renal failure. The kidneys transplanted from water-drinking animals did not develop failure in the salt-drinking animals, but those from salt drinkers did develop failure in the water drinkers. Therefore overhydration of the recipient by continuous salt loading is important in preventing acute tubular necrosis in renal transplantation.

CONTINUOUS salt loading has been shown to protect animals remarkably against acute tubular necrosis from shock, toxins, hæmolysis, or ischæmia (McDonald *et alii,* 1969; Thiel *et alii,* 1970, Silber, 1974). Severe myohæmoglobinuric acute renal failure is produced in normal water-drinking rats by injecting them intramuscularly with 50% glycerol solution, 10 ml/kg. However, in rats which are only allowed to drink 1% NaCl, glycerol injection produces no significant renal failure. No protection is conferred by an acute expansion of plasma volume. Oken's group has shown through elaborate micropuncture studies that acute renal failure is not really a tubular entity, but an angiospastic phenomenon resulting in cessation of glomerular filtration (Oken *et alii,* 1966; Oken *et alii,* 1970). Hollenberg *et alii* (1970) have confirmed in humans through xenon washout studies that "acute tubular necrosis" results from decreased renal cortical perfusions, presumably because of a sustained preglomerular vasoconstriction. Since plasma renin is known to be elevated in acute renal failure, it has been suggested that chronic salt loading protects the kidneys by depleting the body of renin.

By performing renal transplantation between salt-protected and non-protected rats, we hoped to determine if salt affords protection by an extrinsic effect on the host or an intrinsic effect on the kidney itself. This knowledge would be of theoretical and practical importance in renal transplantation.

METHODS

1. Five male DA rats weighing 200 to 300 gm were studied in each experimental group. Members of all groups were given 50% glycerol by intramuscular injection, 10 ml/kg. One group received 10 ml of normal saline intravenously immediately prior to the glycerol injection. Three groups were only allowed 0·9% NaCl to drink for one week, for one month, or for two months. Two groups only received tap water, with no acute or chronic saline load. One of these groups had undergone unilateral nephrectomy prior to glycerol injection. All animals were followed with blood urea level determinations.

2. Each member of a group of five chronic salt-drinking (two months) DA male rats received an

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isogeneic renal allograft from a water-drinking rat along with bilateral nephrectomy. Similarly, five water-drinking DA male rats each received an isogeneic renal allograft from a chronic salt-drinking rat (two months), along with bilateral nephrectomy. After adequate time for recovery from anæsthesia, averaging four hours, each recipient was given 50% glycerol by intramuscular injection, 10 ml/kg. All animals were followed with blood urea level determinations.

Table 1

Blood Urea Level One Day After Glycerol Injection mg/100 ml (S.E. = ±1)

1. Controls

No pretreatment	Chronic salt drinking		
	1 week	1 month	2 months
179·6±10·9	186·8±6·6	98·2±12·1	56·6±17·9

Unilateral nephrectomy beforehand	10 ml intravenous saline
243·6±20·6	185·2±12·3

Renal transplantation was performed with the technique of Fabre *et alii* (1971). A left orthotopic transplant was performed with end-to-end anastomoses of the renal artery and vein and ureter, using 10/0 Nylon sutures (Ethicon, New Jersey, U.S.A.). Ischæmia times were never over 30 minutes.

Results

As can be seen from Table 1, a huge expansion of plasma volume with an acute saline load (equivalent to about 10 litres in the human) gave no protection against renal failure as compared with results in controls. Two months of continued forced saline drinking, however, gave complete protection against renal failure; one month of saline drinking gave partial protection; one week of saline drinking gave no protection. Unilateral nephrectomy significantly elevated the degree of azotæmia ($P<.01$) after glycerol injection, indicating that renal shut-down is not 100% in this model; for otherwise unilateral nephrectomy would make no difference in the degree of the elevation of the blood-urea level.

Table 2

Blood Urea Level One Day After Glycerol Injection mg/100 ml (S.E. = ±1)

2. Glycerol given to recipient after transplant and bilateral nephrectomy

NaCl Kidney→H_2O Rat	H_2O Kidney→NaCl Rat
261·4±15·4	51·4±5·5

Table 2 demonstrates that a kidney removed from a properly salt-protected animal and transplanted into a normal water-drinking animal was no longer protected against acute renal failure. On the other hand, a kidney from a normal unprotected animal, when transplanted into a salt-protected animal, is completely protected against acute renal failure from glycerol injection. Thus the condition of the host, rather than that of the donor, is what protects the kidney against renal failure in this situation.

Discussion

The results of these experiments indicate that the protection afforded by chronic salt loading against acute renal failure is mediated by an effect upon the host rather than upon the kidney itself. If renin depletion alone were important, one would expect the kidney from the normal unprotected animal to be fully susceptible to renal failure even in the protected host. Further, the kidney from the protected animal would be depleted of renin, and therefore would not develop renal failure even in the unprotected host, if renin depletion were the only mechanism of salt protection against acute renal failure. Therefore, the mechanism of protection must be more than renin depletion of the kidney.

In the ischæmic, rather than the myohæmoglobinuric model, we found salt protection to be important in the donor and the recipient, but this could be due to the fact that during the ischæmic period, the kidney is neither in a protected nor an unprotected environment. Furthermore the protection covered only a short duration of ischæmia, under one and a half hours. So although the previous studies may be of interest in renal transplantation, one cannot draw accurate physiological conclusions from them.

A more controlled investigation such as the present one could only be successfully carried out once we had a technique of transplantation with a sufficiently short ischæmia time as not to cause renal damage over and above the glycerol effect. With the microsurgical expertise of this Unit and the previously described technique of renal transplantation (Fabre *et alii*, 1971), it was possible to reduce ischæmia times in this study to levels sufficiently low as to cause no renal failure in salt-drinking, glycerol-injected controls.

These findings suggest that salt loading in the renal transplant recipient may be important in determining the function of the transplanted kidney. We have all noted the odd experience of transplanting two kidneys from one donor into two separate recipients, with one functioning immediately, and the other exhibiting acute tubular necrosis. In our experience the kidney that worked has often been transplanted into an ascitic, poorly dialysed recipient that has not been properly following his directions for restricting salt intake. Furthermore, the kidney that is obviously in acute tubular necrosis 24 hours after transplantation often appeared to be working normally initially. These observations are compatible with a renal vasospastic phenomenon that in some way can be modified by the state of salt balance of the recipient.

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REFERENCES

FABRE, J., LIM, S. H. and MORRIS, P. J. (1971), AUST. N.Z. J. SURG., 41: 69.

HOLLENBERG, N. K., ADAMS, D. F., OKEN, D. E. ABRAMS, H. L. and MERRILL, J. P. (1970), *N. Engl. J. Med.*, 282: 1329.

MCDONALD, F. D., THIEL, G., WILSON, D. R. DIBONA, G. S. and OKEN, D. E. (1969), *Proc. Soc. exp. Biol. (N.Y.)*, 131: 610.

OKEN, D. E., ARCE, M. L. and WILSON, D. R. (1966), *J. clin. Invest.*, 45: 724.

OKEN, D. E., DI BONA, G. F. and MCDONALD, F. D. (1970), *J. clin. Invest.*, 49: 730.

SILBER, S. J. (1974), *Surgery*, 75: 573.

THIEL, G., MCDONALD, F. D. and OKEN, D. E. (1970), *Nephron*, 7: 67.

A Comparison of Cryoprecipitated Plasma and Albumin Solutions for Machine Preservation of Ischæmically Damaged Canine Kidneys[1]

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Four solutions for machine perfusion and preservation of ischæmically damaged canine kidneys were compared. They were (i) canine cryoprecipitated plasma, (ii) canine albumin, (iii) human stable plasma protein, and (iv) human albumin. The results with both canine protein solutions were satisfactory. The solutions containing human protein were found unsuitable for canine kidney preservation. It was concluded that albumin solutions were at least as effective as cryoprecipitate for 24-hour perfusion of ischæmically damaged canine kidneys.

THE major advance which allowed practical development of organ preservation by machine perfusion before allotransplantation was establishment by Belzer *et alii* (1967), that cryoprecipitated plasma was a satisfactory perfusion fluid for prolonged preservation. Since then most groups have used cryoprecipitated plasma for machine preservation of renal allografts. However, preparation of this is generally acknowledged to be troublesome. There is also concern that it may contain human globulins, including antibodies, with the possibility of interaction with the perfused kidneys. An added risk with plasma solutions is the small but significant possibility of transmission of hepatitis. Fears that results of trans-

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