Urinary Acidification in Renal Allografts

G GRAZIANI, A de VECCHI, C PONTICELLI
Ospedale Policlinico, Milan, Italy

Summary
Urinary acid excretion was measured in 35 human renal allograft recipients during the first few days after the onset of diuresis and two months after transplantation. In 16 out of 17 rejection episodes an early significant decrease of renal total net H⁺ excretion was observed. This urinary acidification impairment preceded the serum creatinine increase.

Only few patients showed variations in blood pH and Cl⁻. The acidification defect may be due to the ischaemic changes of rejection.

Early impairment of urinary acidification supports a clinical diagnosis of rejection.

Introduction
The ability to acidify the urine is often impaired in the first few months after kidney transplantation (Gyory et al, 1969; Better et al, 1969). This may be related to several factors such as hyperparathyroidism (Siddiqui and Wilson, 1972), malnutrition (Klahar et al, 1970), and acute tubular necrosis (Better et al, 1970).

Some works have shown that acute rejection may be accompanied by one or more defects in renal hydrogen-ion excretion (Mookerjee et al, 1969; Wilson and Siddiqui, 1973). These reports emphasize the need for further investigation in urine acidification in renal allograft recipients.

We undertook this study to evaluate the capacity of the kidney to acidify the urine in the few weeks following renal transplantation, and particularly during rejection.

PATIENTS AND METHODS
In 15 patients with cadaveric renal transplantation, urinary ammonium, titratable acid, and bicarbonate excretion were measured daily in the first 10 days after
diuresis started. The same parameters were studied, in the next two months, for a period of 7 days, daily or on alternate days, in 35 transplanted patients. During the study all the patients received a free hospital diet. None of them received diuretics or dialysis. None showed urological, gastric or respiratory complications. No signs of urinary infection were present.

Rejection episodes were diagnosed when serum creatinine increased by 25% or more. Other immunological, laboratory, and histological data were obtained to support diagnosis. Blood pH was determined on arterial or capillary blood taken anaerobically.

Collection of 24-hr urine samples was by catheter or spontaneous voiding into flasks containing mineral oil and thymol crystals. Urinary ammonium was determined by the method of Fawcett and Scott (1960). Urine was titrated to blood pH by N/100 NaOH. Urinary bicarbonate was calculated by the Henderson-Hasselbalch equation. Urinary pCO₂ was determined by a radiometer electrode. Total net H⁺ excretion was calculated as the sum of ammonia and titratable acid minus bicarbonate.

Blood Cl⁻ was determined by the method of Cotlove et al (1958) and K⁺ was measured by a flame spectrophotometer.

RESULTS

After the onset of diuresis, total net H⁺ increased progressively up to high and stable levels by about the eighth day (Figure 1.). After this period, in 18 patients without signs of rejection, the percentage variation of total H⁺ excretion ranged between +50% and −40%.

![Figure 1. Mean values of total net H⁺, ammonium and titratable acid excretion in 15 cadaver kidney recipients in the first ten days after the diuresis onset.](image)

During rejection episodes the total H⁺ excretion fell rapidly by more than 40%. This fall preceded an increase in serum creatinine in 16 out of 17 cases (Figure 2).
Figure 2. Percentage variation of total net H⁺ excretion in 18 transplanted patients without symptoms of rejection (above) and 17 cadaver kidney recipients during rejection episodes (below).
Both ammonium and titratable acid excretion defect contributed to the impairment of urinary acidification. The total \( H^+ \) excretion per nephron (\( U_{H^+} + V \cdot \text{creatinine clearance} \)) was significantly reduced during rejection (Figure 3).

![Graph showing blood pH, bicarbonate, chloride, and potassium levels](image)

Figure 3. Variation of blood pH, bicarbonates, chloride and potassium during rejection episodes. (0 day = when serum creatinine increased more than 25%.)

During rejection episodes blood pH decreased in 7 out of 13 patients and serum bicarbonate decreased in 9 out of 10 patients. In 3 out of 10 patients blood Cl\(^-\) increased. No variation was observed in blood K\(^+\) (Figure 4).

![Graph showing \( U_{H^+} \cdot V / \text{Cr:Cl} \) and pEq/ml](image)

Figure 4. Variation of total net \( H^+ \) excretion ‘per nephron’ in 17 transplanted patients during rejection episodes.

**DISCUSSION**

Our data show that, in the first few days after diuresis onset, urinary acidification gradually increased. This may be due to several causes; osmotic diuresis

After about eight days, renal H⁺ excretion ranges between 60 and 80μEq/min. This corresponds to the mean daily endogenous acid production (Richet et al, 1966). The ammonium excretion was greater than 40 μEq/min, and the titratable acid excretion was greater than 25 μEq/min. Similar values are observed in normal kidney after an NH₄ Cl load (Wrong and Davies, 1959).

On a standard diet and if no other factors interfere with the acid-base balance, this amount fluctuates between +50% and −40%. During acute rejection episodes, there is a fall in renal H⁺ excretion. In some patients, however, no simultaneous decrease of blood pH was observed.

This is probably due to the fact that intra- and extra-cellular buffers may neutralize excess endogenous H⁺ production.

A defect in H⁺ excretion was evident in 16 out of 17 patients before serum creatinine increased. Therefore, this alteration should be considered a specific consequence of rejection; it is not — at least initially — caused by a decrease of glomerular filtration. This is confirmed by the finding that H⁺ excretion ‘per nephron’ is also reduced.

Some hypotheses may be proposed to explain how rejection causes such a rapid rejection in acid excretion.

Distal renal tubular acidosis has been reported to occur in a variety of autoimmune diseases, such as systemic lupus erythematosus (Tu and Shearn, 1967), autoimmune thyroiditis (Mason and Golding, 1970), and Sjogren’s syndrome (Shioj et al, 1970).

In these situations the acidification defect appears to be related to mononuclear cell infiltration of the kidney interstitium (Mahieu et al, 1972). Since in acute rejection an interstitial and lymphoid cell reaction is often observed (Kountz et al, 1963; Porter et al, 1964) the defect in acidification may be attributed to cell-mediated immunity.

There is strong evidence that ischaemia follows the immunological injury (Kountz et al, 1965; Retik et al, 1967).

Changes in the cortical component of blood flow occur early, preceding changes in total renal flow (Retik et al, 1967; Hollemberg et al, 1968). It is known that the glomeruli have an autoregulation system which may protect them against ischaemia; this mechanism has not yet been shown in the peritubular circulation. Thus it is possible to surmise that cortical ischaemia causes functional impairment of the tubules before involving the glomeruli. There is also evidence, however, that immunological deposits may be seen on the basement membranes of renal tubules during transplantation and in acute rejection (Andres et al, 1970; Williams et al, 1967). Our own experience confirms that deposits of complement and immunoglobulins on the tubular basement membranes can be observed in some cases of acute rejection (Figure 5).
Figure 5. Deposits of complement and immunoglobins along tubular basement membranes during an acute rejection episode (x300).

These findings suggest that both cellular and humoral immunity may be involved in the aetiology of urinary acidification defect during acute rejection.

In fact, in serially examined transplant patients a rapid fall of urinary acidification may be interpreted as an early sign of acute rejection. Obviously, other variables which can influence renal acidification ability – diuretics or dialysis, severe hyper- or hypokalaemia, gastric or respiratory lesions – must be excluded.

References

Andres, G A, Accini, L and Hsu, K C (1970) Laboratory Investigation, 22, 588
Graziani, G, De Vecchi, A, Rivolta, E, Limido, D and Ponticelli, C (1973) Minerva Nefrologica, 20, 1
Mahieu, P, Dardenne, M and Bach, G F (1972) American Journal of Medicine, 53, 185

282
Open Discussion

CHAIRMAN As far as I could see from your slides you only showed us total hydrogen ion excretion in the urine. I may be mistaken, but would you kindly give us, or restate, the total hydrogen ion/creatinine clearance ratio?
PONTICELLI We have measured total hydrogen ion excretion/creatinine clearance ratio. We have observed that this ratio decreases during rejection episodes.

S MASSRY (Los Angeles) In discussing renal tubular acidosis after renal transplant, I think one should differentiate between the two types of renal acidosis, namely proximal and distal. The distal type is still a puzzle to me but it could be related to acute tubular necrosis, or immunological problems, giving rise to decreased acid excretion. The proximal type is commonest after a renal transplant, and one finds a fall in blood bicarbonate, but there is no distal tubular problem. You suggested hyperparathyroidism as the most likely cause, but unfortunately experimental data no longer support the idea that parathyroid hormone in the usual concentrations we see in the blood really reduce Tm bicarbonate. I would like to propose to you another possibility, now substantiated by experimental work, phosphate depletion. Phosphate depletion with hypophosphataemia is associated with decreased Tm bicarbonate, and most transplant patients have hypophosphataemia after transplantation, with hyperchloaemic acidosis. Dr Moorhead presented data a week ago on patients with hyperchloraemic acidosis and reduced Tm bicarbonates months after transplant when his patients had hypophosphataemia. Did you measure the phosphate in the blood, and how did you relate your findings to this possibility?
PONTICELLI We have observed a relative hypophosphataemia in some patients, but we did not correlate this with Tm bicarbonate.