Immune potential in human uraemia

2 Changes after regular haemodialysis therapy

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SYNOPSIS Parameters of both humoral and cellular immune potential were measured in a group of patients with severe renal failure before and after three months' regular haemodialysis therapy. Evidence is presented of improvement in cellular immune potential and of a tendency of the response of lymphocytes to PHA to return to normal. No improvement in humoral responsiveness was demonstrable, and it is suggested that uraemic patients on regular haemodialysis may have an impaired capacity to establish new immunological memory.

We have shown that both humoral and cellular immune-potential are progressively diminished with decreasing glomerular filtration rate (GFR) (Byron *et al.*, 1976) and that the degree of renal failure at which the impairment becomes readily detectable corresponds to a GFR of approximately 10 ml/min. In our hands, maintenance of a patient with essentially no renal function on regular haemodialysis corresponds to an effective average GFR of 10-15 ml/min. These studies were carried out to investigate whether improvement in immune potential resulted from regular haemodialysis, as might be predicted by the increase in effective average GFR.

Material and methods

The seven patients investigated were all in severe renal failure. Five were female and two male, and the aetiology of the renal disease included polycystic kidneys (2), chronic pyelonephritis (2), glomerulonephritis (1), postpartum cortical necrosis (1), and unknown causation (1). The mean GFR of the group was 2.3 ml/min and the mean serum creatinine was 15.5 mg/100 ml. None of the patients was taking drugs likely to interfere with immune potential.

Methods used in the investigation of cellular and humoral potential were those we have used previously (Byron *et al.*, 1976) except that contact sensitization to dinitrochlorobenzene was not included. Humoral immune potential was quantitated by the measurement of antibody titres before and 17 days after immunization with TABT vaccine. Cell-mediated potential was investigated by the determination of 48-hour skin induration after injection of a bank of commonly experienced antigens and by

the *in vitro* blastogenic responses to tuberculin, *Candida albicans*, and phytohaemagglutinin (PHA).

The 19-day protocol was carried out on each patient before the institution of regular haemodialysis and again after at least three months' regular haemodialysis. The results, before and after treatment, were compared using the paired *t*-test. Titres were converted to log₁₀ for all comparisons. Blastogenic responses were compared as blastogenic indices:

$$\begin{aligned} & \text{mM } ^3\text{H TdR} \times 10^{10}/10^6 \text{ lymphocytes in test} \\ & - \text{mM } ^3\text{H TdR} \times 10^{10}/10^6 \text{ lymphocytes in} \\ & \quad \text{unstimulated culture.} \end{aligned}$$

Results

The results of the antibody responses to TABT are presented in table I. Most of the subjects failed to produce antibody to *Salmonella paratyphi* A antigens either before or after haemodialysis, and therefore these results are not included. Only with respect to *Salm. typhi* 'H' was a significantly greater amount of antibody present before the post-dialysis immunization than at the start of the protocol ($P < 0.001$). Thus, although a significant rise in titre to *Salmonella typhi* and *paratyphi* B antigens and tetanus toxoid occurred after the initial immunization with TABT, only the response to *Salm. typhi* 'H' persisted during the period on haemodialysis. Comparing the 17-day samples of serum before and after haemodialysis, significant differences were not demonstrated. The titres achieved in response to each of the two doses were very similar although it might have been expected that the second of the two doses of vaccine would have induced a considerably augmented response.

Before haemodialysis the mean of the sums of 48-hour skin responses to the four antigens given

Antigen	Pre-Haemodialysis		Post-Haemodialysis	
	Day 1	Day 17	Day 1	Day 17
Tetanus toxoid	ND	1.59 (1.29)	0.99 (0.59)	1.98 (1.01)
<i>Salm. typhi</i> 'O'	0.86 (0.64)	1.62 (1.01)	1.19 (0.49)	1.76 (0.48)
<i>Salm. typhi</i> 'H'	ND	2.50 (0.62)	1.63 (0.58)	2.38 (0.42)
<i>Salm. paratyphi</i> B 'O'	0.80 (0.45)	1.43 (0.79)	1.17 (0.64)	1.62 (0.81)
<i>Salm. paratyphi</i> B 'H'	ND	1.06 (1.04)	0.62 (0.61)	1.03 (0.80)

Table I Comparison of responses to TABT before and after haemodialysis

Mean and standard deviations (in parenthesis) of log₁₀ titres on day of immunization and on day 17.

ND = none detected

Mitogen	Pre-haemodialysis	Post-haemodialysis
PPD 250 u	27.4	59.3
PPD400 u	39.4	76.5
PHA 5 µl	168.0	71.7
Nil (unstimulated)	3.8	12.4

Table II Mean blastogenic indices after 120 hours in culture of lymphocytes taken before and after establishing haemodialysis

intradermally was 26.9 mm, ie, below the normal range of 30-60 mm. After three months' haemodialysis this parameter of cell-mediated reactivity rose to 32.9 mm. The increase is, however, not statistically significant ($P > 0.1$).

Changes were demonstrated in the *in vitro* reactivity of lymphocytes when pre- and post-haemodialysis values were compared. These are presented in table II. Our previous results had shown a trend of diminishing blastogenic response to PPD with decreasing GFR (Byron *et al*, 1976). This trend is to some extent reversed after haemodialysis with a response which is approaching the normal. The change is, however, not significant ($P > 0.2$).

We have also previously demonstrated a slower response to PHA mitogenesis in patients with low GFRs so that comparison of results after periods of incubation longer than 72 hours reveals significantly greater uptake of thymidine in the patients with severe renal failure than in normal subjects. This trend is also reversed by haemodialysis. A significant reduction ($P < 0.05$) in 120-hour PHA-induced blastogenesis was found when pre- and post-haemodialysis results were compared. Interpretation of this change is made more difficult by a non-significant ($P > 0.1$) increase in the uptake of thymidine by unstimulated lymphocytes after haemodialysis. The changes observed probably represent a combination of increased control uptake of thymidine together with a tendency to return to a more normal timing of peak transformation.

Discussion

Patients in severe renal failure show impaired protein synthesis which is restored by adequate regular dialysis therapy (Coles *et al*, 1970). The haemopoietic system shares the effects of these changes in protein metabolism, for example, haemoglobin production improves on regular dialysis. However, abnormalities in the plasma aminogram persist despite regular dialysis, suggesting remaining defects in protein metabolism, and while the patient becomes able to lead an active working life, residual metabolic impairment (for example, in bone metabolism and carbohydrate handling) is detectable.

Our results show that regular dialysis therapy (RDT) for three months improves the cellular immune response as measured *in vitro* in response to tuberculin, and that there is a tendency for a return towards normal of PHA responsiveness. *In vivo* delayed hypersensitivity responses to common antigens also improved. Significant improvement in the depressed humoral response to the antigens used was not observed. This implies an even greater deficit than the low titres themselves reveal as the patients would have been expected to produce a secondary response in view of the primary dose of antigen given before dialysis was started. This suggests that, despite RDT, the patient in severe uraemia has an impaired capacity for the establishment of new immunological memory at least in the humoral compartment.

We did not carry out further testing after RDT for periods longer than three months and so cannot be sure that the values obtained represent a plateau beyond which the patient cannot improve. Clinical evidence (of occult tuberculosis and persisting viral infection, eg, hepatitis B) suggests that, as in other systems, there are long-term persisting defects in immunity in RDT patients. The precise localization of defects—macrophage function, immunological memory, effective cellular and humoral immunity—has not been documented systematically; it seems probable that multiple defects arise and that the changes may not be uniform in all patients. For example, while there is no doubt that cellular and humoral response to a renal allograft in a RDT patient can be both rapid and effective, recent evidence suggests that not all such patients may possess the capacity to recognize a non-identical allograft and mount a response. Using mixed leucocyte reaction (MLR) with non-identical donor lymphocytes, RDT patients have been divided into non, poor or good responders, and rejection of the subsequent renal allograft has been shown to relate to this classification, non or poor responders retaining the graft, good responders rejecting it (Cochrum

et al, 1974). Subsequent to this investigation all seven patients have received cadaver renal allografts. In five the grafts are functioning well after periods in excess of one year. One patient required a transplant nephrectomy because of rejection, and one patient died of sepsis and gastrointestinal haemorrhage. We could detect no relationship between the parameters of immunological function which we measured and the fate of the renal allograft in this small series. Thus either the number of patients investigated is too small to reveal a relationship or changes in MLR response represent a further abnormality of

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immune potential in uraemia unrelated to those we have investigated.

References

- Byron, P. R., Mallick, N. P., and Taylor, G. (1976). Immune potential in human uraemia 1 Relationship of glomerular filtration rate to the depression of immune potential. *J. clin. Path.*, **29**, 00-00.
- Cochrum, K. C., Salvatierra, O., and Belzer, F. O. (1974). Correlation between M.L.C. stimulation and graft survival in living related and cadaver transplants. *Ann. Surg.*, **180**, 617.
- Coles, G. A., Peters, D. K., and Jones, J. H. (1970). Albumin metabolism in chronic renal failure. *Clin. Sci.*, **39**, 423.