Effect of Haemodialysis on Immune Response

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Summary

Uraemia depresses immune response by altering cellular reactivity to mitogenic and antigenic stimulation.

One might expect that amelioration of uraemia by dialysis would improve immune responses. We have investigated the effect of haemodialysis on in-vitro parameters of cellular immunity. Our data suggest that haemodialysis leads to loss of a factor or factors from both uraemic and normal plasma essential for DNA synthesis. Our data do not suggest that measurements of cellular immunity are useful in monitoring adequacy of haemodialysis in chronic uraemic patients.

Introduction

Uraemia is believed to depress immune response, alter cellular reactivity to mitogens and to antigens and impair production of humoral antibodies (Wilson and Kirkpatrick, 1965; Kirkpatrick and Wilson, 1964; Lawrence, 1965). Clinically this phenomenon has been correlated with absence of skin sensitivity to antigens and an improved homograft survival in uraemic patients (Couch and Murray, 1957; Hume and Merrill, 1955; Lawrence, 1965).

The presence of a dialysable factor in uraemic serum that is responsible for the suppression of cellular immunity has been suggested (Newberry and Sanford, 1971; Silk, 1965). Reversal of such depression of cellular immunity may then be expected following adequate haemodialysis. We have studied the cellular response to mitogen and antigens measured by $^{14}$C thymidine incorporation in DNA of transforming lymphocytes in vitro before and following adequate haemodialysis.

MATERIALS AND METHODS

The macro method for lymphocyte transformation has been reported fully elsewhere (Valentine, 1971). The following concentrations of mitogen and antigens were used: phytohaemagglutinin (PHA-M) 0.1 ml per tube (Difco); purified protein derivative (PPD) 250 units per tube (Merck, Sharp and Dohme);
streptokinase-streptodornase (SKSD) 40 units per tube (Lederle).

The micro method for lymphocyte transformation (Valentine, 1974) has an advantage over the macro method. The number of lymphocytes used is smaller (5 × 10⁵/ml) and consequently the volume of blood and amount of antigen used are reduced to one-twentieth of that used in the macro method.

Seven patients with chronic uraemia were studied while undergoing haemodialysis for the first time. Methyl cellulose acetate hollow-fibre dialysers were used in all patients with a standard dialysate composition (Na 132 mEq/K 0.2 mg%) Ca 8 mg%, 1.5 mg% glucose 200 mg%). Blood samples for in vitro lymphocyte transformation as well as chemistries were taken before and after haemodialysis. The macro method for lymphocyte transformation was used. Eight studies were performed in seven patients.

In four ‘stable’ uraemic patients who were not dialysed, lymphocyte transformation was studied using autologous (uraemic) plasma and heterologous (non-uraemic) plasma. Studies were done with PHA and PPD and the micro method was used in all.

To study the effect of dialysis on lymphocyte transformation in normal, non-uraemic blood, 200 ml of venous blood from normal donors was dialysed in a ‘closed circuit’ using methyl cellulose acetate hollow-fibre dialysers and standard dialysate. Blood samples were drawn at regular intervals and lymphocyte transformation was studied by the micro method. Experiments were carried out in two sets, one utilising autologous dialysed plasma and in the other, heterologous (non-dialysed) pooled AB+ plasma.

RESULTS

The degree of uraemia of patients varied and we have observed no correlation between uraemic indices and cellular response before haemodialysis. Incorporation of ¹⁴C thymidine following exposure to PHA in uraemic patients in our series was comparable to our normal controls, although there were three patients who exhibited low responses to PHA as well as PPD and SKSD (Figure 1).

Washed uraemic lymphocytes resuspended in medium containing heterologous, non-uraemic, plasma showed consistently greater response with PHA and PPD compared to when suspended in media containing autologous, uraemic, plasma (Figure 2 and 3).

After six hours of haemodialysis, a decrease in ¹⁴C thymidine incorporation was observed to our surprise in a majority of uraemic patients with PHA, SKSD and PPD (Figure 4). Here, washed lymphocytes were resuspended in medium containing autologous dialysed, plasma.

We have also examined some of the variables that might play a role in the causation of this dialysis-induced depression of cellular immunity in vitro.
Figure 1. Response to PHA of lymphocytes from uraemic patients prior to dialysis and in normal subjects. (Macro method.)

Haemodialysis produced lymphopenia (Figure 5) with relative increase in granulocytes. Since the number of lymphocytes used in each of these experiments is always kept constant, the decrease in $^{14}$C thymidine incorporation produced by dialysis cannot be attributed to quantitative changes in lymphocyte population.

Normal dialysed lymphocytes when suspended in heterologous (non-dialysed) pooled AB+ plasma retained their capacity to respond to PHA, PPD and SKSD even after two hours of 'closed circuit' dialysis, whereas the lymphocytes suspended in media containing autologous (dialysed) plasma completely lost their ability to respond to mitogen within thirty minutes. This suggests that the depression of stimulation seen following dialysis results primarily from the loss of a plasma factor during dialysis rather than from an alteration in the population of cells (Figure 6).
Figure 2. Response to PHA of lymphocytes from uraemic patients in autologous and heterologous plasma. (Micro method.)

Figure 3. Response to PPD of lymphocytes from uraemic patients in autologous and heterologous plasma. (Micro method.)
DISCUSSION

Lymphocytes from uraemic patients and normal controls in our series had comparable response to PHA. Similar observations were reported by Kasakura and Lowenstein (1967), and also Daniels and Remmers, et al (1970). However, our study as well as those of Newberry and Sanford (1971) and Silk (1965), support the hypothesis that a factor in uraemic plasma inhibits lymphocyte transformation.

Haemodialysis, although improving uraemic indices, appears to diminish lymphocytic response to antigens. In addition to producing lymphopenia, dialysis seems to impair the ability of dialysed plasma to act as a medium in which cells can respond to antigenic stimulation. Similar results have been published by Nelson and Penrose (1972). They postulated that this depression in cellular response following haemodialysis is due to a non-dialysable factor which actively inhibits DNA-synthesis in lymphocytes.
Figure 5. Absolute lymphocyte count in uraemic patients before and after dialysis.

Figure 6. Response to PHA of lymphocytes from normal subjects undergoing 'closed circuit' dialysis. (Micro method.)
Our experiments suggest that haemodialysis leads to loss of a factor or factors from normal and uraemic plasma which is essential for DNA synthesis. Based on the present understanding of permeability characteristics of cellulose-acetate membrane this factor or factors is likely to be of low molecular weight.

Our data suggest that in vitro assessment of cellular immunity is not helpful in evaluating the adequacy of haemodialysis in chronic uraemic patients.

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