

## **INTERLEUKIN-1 IN HYPERSENSITIVITY REACTIONS DURING HAEMODIALYSIS**

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### **Summary**

Interleukin-1 (IL-1) produced by peripheral monocytes was studied in 23 dialysis patients treated with cuprophan and polyacrylonitrile membranes. IL-1 activity was significantly higher in cuprophan patients than in polyacrylonitrile patients ( $p < 0.02$ ). In five patients studied during allergic reactions the IL-1 activity was reduced compared with values before treatment ( $p < 0.02$ ) with IgE anti-ethylene oxide. The T-lymphocyte response to IL-1 from normal monocytes did not differ in both groups.

The authors conclude that chronic activation of blood monocytes induced by cuprophan membranes may be involved in hypersensitivity reactions during dialysis sessions.

### **Introduction**

Severe reactions resembling anaphylaxis occur in a variety of situations [1-4] during haemodialysis. They range from mild symptoms to life-threatening bronchospasm, hypotension and cardiopulmonary collapse. The substances responsible for this phenomenon seem to be a component of the cellulose membrane or additional compounds used in capillary dialysers [4]. Reaction to ethylene oxide gas, commonly used in sterilization, has also been hypothesized [5].

Complement activation during haemodialysis sessions has been reported and correlated with several biological activities, such as leukoaggregation and pulmonary vascular leukosequestration [6], their intensity depending on the type of dialysis membrane used and whether or not filters were reused. However, the debate on the mechanism responsible for this phenomenon continues.

This study describes the mononuclear phagocytic cell activation observed during haemodialysis with different dialysis membranes and its role in hypersensitivity reactions.

## Patients and methods

**Patients** Twenty-three patients (13 male, 10 female), mean age  $58.16 \pm 12.8$  years, on stable chronic haemodialysis with new dialysers for at least three months, were studied. Sixteen patients were treated routinely with cuprophan membranes (CF 12-11, CF 15-11 Travenol Laboratories or Gambro 5N, Gambro AB) and seven with polyacrylonitrile membrane devices (H 1210, H 1207, H Hospital). Before use, all dialysers were rinsed with two litres of sterile saline.

Twenty patients with end-stage renal disease (mean creatinine clearance =  $16 \pm 4$  ml/min) and 10 healthy members of the hospital staff served as controls for the LAF assay and for IL-1 production.

**Generation of IL-1** Mononuclear cells, separated in a Ficoll-Hypaque density gradient were counted and corrected up to  $2.5 \times 10^6$  cells/ml in RPMI containing 5% fetal calf serum (FC-Eurobio) [7].

Two millilitres of this cell suspension were dispensed into wells of Nunc plates and incubated in 5% CO<sub>2</sub> at 37°C. After 8–12 hours the non-adherent cells were removed and the adherent cells were stimulated with *E Coli* lipopolysaccharide (1 µg/well). The plates were incubated in 5% CO<sub>2</sub> in air at 37°C for 48 hours and then the supernatants were collected and stored at -20°C until assayed.

**IL-1 assay (LAF assay)** A 100 µl culture of  $8 \times 10^6$  C3H-H mouse thymocytes were plated into flat-bottomed microtitre plates. IL-1 preparations were added in triplicate at 1:3, 1:9, 1:27 dilutions with PHA-M (DIFCO), 1:150 dilution. After incubation in 5% CO<sub>2</sub> in air at 37°C for 48 hours the plates were pulsed with 5H-thymidine, at 2 µCi/well and harvested 18–20 hours later.

Results are expressed in c.p.m. and IL-1 activity was evaluated by stimulation

$$\text{index} = \frac{\text{c.p.m. PHA with IL-1}}{\text{c.p.m. PHA without IL-1}}$$

**Assay for T-lymphocyte response to IL-1** This test was evaluated by induction of stable E-rosette forming cells (active E-RFC) with IL-1 [8].

Lymphocytes ( $4 \times 10^6$  cells/ml) were incubated in triplicate with IL-1 from normal subjects at 1:3, 1:9 dilutions. After 60 hours of culture, the cells were harvested, washed twice with RPMI 1640, incubated with an equal volume of sheep red blood cells (SRBC—Sclavo 5%) ( $8 \times 10^7$  cells/ml) and centrifuged at 200g for five minutes. The cells were gently resuspended and the percentage of E-RFC was determined. Data are presented as  $\Delta\% = \%$  of active E-RFC formed in the presence of IL-1 minus the percentage of active E-RFC formed without IL-1.

**IgE** Total IgE (PRIST assay Pharmacia) and IgE anti-ethylene oxide (RAST test Pharmacia) were studied.

**Statistics** Statistical analysis was made by Student's 't' test for paired and unpaired data.

## Results

*Production of IL-1 from cells of dialysis patients and controls* IL-1 activity in the supernatants of cells isolated from end-stage renal dialysis patients differed significantly ( $p < 0.01$ ) from that in normal subjects.

Figure 1 shows the variation of IL-1 at the beginning and at the end of the dialysis with both cuprophane and polyacrylonitrile membranes.

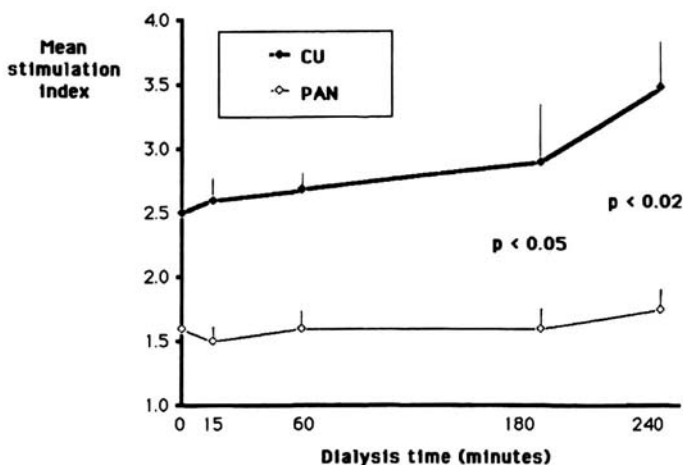


Figure 1. IL-1 activity, expressed as mean stimulation index, in patients treated with cuprophane (CU) and polyacrylonitrile (PAN) membranes

IL-1 production from peripheral monocytes was significantly higher in patients treated with cuprophane membranes than in those treated with polyacrylonitrile membranes.

In five cases studied during an allergic reaction the IL-1 activity was very low compared with values before treatment (stimulation index =  $1.01 \pm 0.02$  versus  $2 \pm 0.02$ ,  $p < 0.02$ ). In three out of four cases IgE anti-ethylene oxide antibodies were also found.

*Response to IL-1* The response of T-lymphocytes from dialysis patients to IL-1 from normal subjects did not significantly differ in patients treated with cuprophane and polyacrylonitrile membranes ( $\Delta\% = 29.4 \pm 5$  and  $26.3 \pm 3.3$ ).

## Discussion

In this study we reported that in vitro differences in the IL-1 production from monocytes of haemodialysis patients corresponded to the different membranes used. When the blood contacted the dialyser cuprophane membrane monocytes, became activated with the consequent release of IL-1; whereas, dialysers containing polyacrylonitrile membrane promoted very little monocyte activation and correspondingly failed to induce IL-1 production.

Transient complement activation and leucopenia have been described during haemodialysis with cuprophane membranes, but only slight activation was seen when polyacrylonitrile membrane was used, because of its limited capacity to bind complement fragments [6]. This phenomenon might be responsible for the monocyte activation found in our patients, because mononuclear phagocytes have been reported to have receptors for  $C_{5a}$  which induce IL-1 production and release [10].

Monocytes of patients dialysed by cuprophane membranes seem to be subjected to chronic activation and in particular situations may release large amounts of IL-1 which may cause hypersensitivity reactions via prostaglandin and leukotriene syntheses.

These results are stimulating but they require further confirmation, particularly with regard to the quantity of the plasma IL-1 levels during haemodialysis.

## References

- 1 Ogden DA. *N Engl J Med* 1980; 302: 1262
- 2 Nicholls AJ, Platts MM. *Br Med J* 1982; 285: 1607
- 3 Key J, Nahmias M, Acchiardo S. *Am J Kid Dis* 1983; 2: 664
- 4 Ing TS, Daugirdas JT, Popli S et al. *Int J Artif Organs* 1983; 6: 235
- 5 Hakim RM, Breillatt J, Lazarus JM et al. *N Engl J Med* 1984; 4: 878
- 6 Chenoweth DE, Cheung AK, Henderson LW. *Kidney Int* 1983; 24: 764
- 7 Dinarello CA. *N Engl J Med* 1984; 311: 1413
- 8 Giacchino F, Giraud G, Pozzato M et al. *Transplant Clin Immunol* 1985; 17: in press
- 9 Dinarello CA. *Rev Infect Diseases* 1984; 6: 51
- 10 Dinarello CA. *Blood Purification* 1983; 1: 197