Detection of Circulating Immune Complexes in Patients on Hemodialysis by a New Immunoassay

T.	Toshio	Miyata ^a
R.	Reiko	Inagi ^b
A.	Akiei	Hamano ^c
I.	Izumi	Nakashima ^b
K.	Kenji	Maeda ^a

^aDepartment of Internal Medicine, The Branch Hospital, and ^bDepartment of Immunology, Nagoya University School of Medicine, Nagoya, and ^cCentral Research Laboratory, Nissui Pharmaceutical Co., Ltd., Ibaraki, Japan

Dr. K. Maeda, Department of Internal Medicine, The Branch Hospital, Nagoya University School of Medicine, 1-1-20, Daikominami, Higashi-ku, Nagoya 461 (Japan)

Dear Sir,

We measured circulating immune complexes (CIC) in patients on hemodialysis by a new, simple and sensitive sandwich immunoassay using mouse monoclonal IgG rheumatoid factor (mlgGRF) specifically reactive with immune-complexed human IgG. mlgGRF was raised from the hybridoma of MRL/lpr mouse spleen cells and NS-1 myeloma cells. Screening was by solid-phase enzyme-linked immunosorbent assay (ELISA) using heat-aggregated human IgG. The sensitivity of this assay system was very high (approximately 0.5 µg/ml of heat-aggregated human IgG equivalent). Particularly, this assay had higher sensitivity for detection of intermediate CIC (MW < 1,000,000) than other assay methods. This was confirmed by comparing the reactivity of mlgGRF with fractions of immune complexes after gel filtration chromatography. Moreover, almost no difference in result between serum and heparinized plasma was observed.

We measured fresh serum samples from 15 healthy individuals and 60 patients on hemodialysis for end-stage renal failure due to chronic glomerulonephritis. As shown in figure 1, the amount of CIC in patients was estimated as 2.05 ± 1.18 µg/ml (mean value $\pm$ SD), while that in healthy individuals was 0.91 ± 0.39 µg/ml. The difference of these values between patients and healthy individuals was statistically significant by an unpaired Student's t test ($p < 0.01$). The 95% confidence limit for normal serum in this assay was 1.69 µg/ml. Hence, about 43% of patients showed positive CIC in the serum.

Until now, some researchers have reported that CIC were detected with a high incidence in patients on maintenance hemodialysis [1] and glomerulonephritis [2], by the method of the Raji cell radioimmunoassay [3], the

Patient (n = 60)

Normal (n=15)

Fig. 1. CIC detected in the sera of patients on hemodialysis and healthy individuals. Horizontal bars indicate the mean $\pm$ SD in each group.

radiolabeled Clq binding assay [4], or the microcomplement consumption test [5]. These indirect assays are based on the immunochemical properties of immune complexes, and each has its special reactivity and sensitivity. Our present result by a new direct immunoassay seemed confirmative to the earlier observations. The value of percentage of patients with positive CIC in this study was higher than that in some of the earlier studies, which might be due to the high sensitivity for detection of intermediate CIC.

Immune complexes have been thought to have various effects on immune response and play a role in the patho-

Detection of CIC in Patients on Hemodialysis by a New Immunoassay

245

Zubler RH, Lange G, Lambert PH, et al: Detection of immune complexes in unheated sera by a modified ¹²⁵I-Clq binding test. Effect of heating on the binding of Clq by immune complexes and application of the test to systemic lupus erythematosus. *J Immunol* 1976;116:232–235.

Mowbray JF, Hoffbrand AV, Holborow EJ, et al: Circulating immune complexes in dermatitis herpetiformis. *Lancet* 1973;i: 400–402.

genesis of some renal diseases. A recent study reported 4 that the presence of CIC in regularly dialyzed patients was associated with prolonged graft survival in renal transplantation [1]. We think that this new, sensitive immunoassay would be useful for detection of CIC, particularly intermediate CIC.

References

Perez GO, Glasson P, Favre H, et al: Circulating immune complexes in regularly dialyzed patients with chronic renal failure. *Am J Nephrol* 1984;4:215–221.

Woodroffe AJ, Border W, Theofilopoulos AN, et al: Detection of circulating immune complexes in patients with glomerulonephritis. *Kidney Int* 1977;12:268–278.

Theofilopoulos AN, Wilson CB, Dixon FJ: The Raji cell radioimmune assay for detecting immune complexes in human sera. *J Clin Invest* 1976;57:169–182.