Clin. exp. Immunol. (1988) **72**, 55–59

Abnormal clearance of soluble aggregates of human immunoglobulin G in patients with systemic lupus erythematosus

S. LOBATTO, M.R. DAHA, F.C. BREEDVELD*, E.K.J. PAUWELS†, JEANNETTE H. EVERS-SCHOUTEN, A.A. VOETMAN‡, A. CATS* & L.A. VAN ES *Department of Nephrology, *Department of Rheumatology, and †Department of Nuclear Medicine, University Hospital, Leiden, and ‡Central Laboratory of The Netherlands Red Cross Blood Transfusion Service, Amsterdam, The Netherlands*

(Accepted for publication 28 October 1987)

SUMMARY

In the present study, we tested mononuclear phagocyte system function in nine healthy controls and 15 SLE patients with complement activating ^{123}I-labelled aggregates of human IgG (AIgG). Clearance half-time of AIgG was 26 ± 8 min in controls, compared to 58 ± 27 min in patients ($P < 0.005$). Binding of AIgG to erythrocytes was significantly lower in patients, 9.3 ± 8.1 vs $24 \pm 20\%$ ($P < 0.05$). The increase of C3a-levels in plasma was significantly lower in patients than in controls ($P < 0.05$ at 3 and 8 min), suggesting less complement activation. Liver and spleen uptake of ^{123}I-AIgG was measured with a gamma camera and expressed as liver/spleen uptake ratios. In patients, the liver/spleen uptake ratios were significantly higher than in controls at 15 min, 3.8 ± 2.0 vs 2.31 ± 0.7 ($P < 0.05$), due to less splenic uptake of AIgG. Correlations between clearance half-time or liver/spleen uptake ratios and immune complex levels or disease activity were not found. This study indicates that clearance of soluble AIgG is abnormal in patients with SLE, due to decreased splenic uptake of AIgG.

Keywords IgG aggregates systemic lupus erythematosus MPS function circulating immune complexes

INTRODUCTION

The mononuclear phagocyte system plays an important role in the removal of immune complexes from the circulation. Studies of mononuclear phagocyte system function using autologous erythrocytes coated with anti-rhesus IgG (E.IgG) have been performed in a variety of diseases (Frank, 1983; Kimberly & Ralph, 1983). In systemic lupus erythematosus (SLE), the clearance half-times ($t_{\frac{1}{2}}$) of E.IgG were found to correlate with both circulating immune complex levels and with disease activity (Frank *et al.*, 1979). In addition, a subsequent longitudinal study by the same group demonstrated that $t_{\frac{1}{2}}$ of E.IgG became shorter as disease activity decreased in individual patients (Hamburger *et al.*, 1982). Several other studies in patients with SLE failed to demonstrate a correlation between $t_{\frac{1}{2}}$ of E.IgG and circulating immune complex levels (Kimberly *et al.*, 1983; Parris *et al.*, 1982; Van der Woude *et al.*, 1984). In most other diseases studied, $t_{\frac{1}{2}}$ of E.IgG did not correlate with disease activity or circulating immune complex levels as determined with the C1q-binding assay.

Correspondence: S. Lobatto MD, Department of Nephrology, University Hospital Leiden, PO Box 9600, 2300 RC Leiden, The Netherlands.

In healthy volunteers and in patients with various immune complex diseases, E.IgG have been shown largely to be removed from the circulation by the spleen (Frank, 1983; Kimberly & Ralph, 1983), while soluble immune complexes in animals are cleared predominantly by the liver (Mannik & Arend, 1971; Veerhuis *et al.*, 1986).

We have recently developed a new method for measuring mononuclear phagocyte system function in humans using soluble aggregates of human immunoglobulin G (AIgG) (Lobatto *et al.*, 1987). A significant advantage of this probe is that AIgG bear a closer resemblance to soluble immune complexes. Therefore, their use may be helpful in giving insight into the mechanisms of immune complex clearance, thereby allowing the establishment of a relationship between mononuclear phagocyte system function and disease activity or circulating immune complex levels.

MATERIALS AND METHODS

Procurement of aggregates of immunoglobulin G

AIgG were produced in the Central Laboratory of the Netherlands Red Cross Blood Transfusion Service, as described by Lobatto *et al.* (1987).

Radiolabelling of AIgG

On the day of the study, one sample of AIgG was thawed, and AIgG was radiolabelled with Na^{123}I, as described by Lobatto *et al.* (1987). The total amount of radioactivity administered to each volunteer or patient was approximately 3·7 MBq (100 μCi).

Selection of healthy controls and patients with SLE

The study protocol was approved by the Ethical Committee of the Leiden University Hospital. Healthy controls and patients participated in the study after informed consent was given. Controls were screened for absence of disease. Patients in whom SLE had been diagnosed based on the revised 1982 ARA criteria for SLE (Tan *et al.*, 1982) were selected from the in-patient and out-patient Rheumatology departments. Disease activity was scored on the day of the study according to a previously described method (Valentijn *et al.*, 1985).

Special attention was given to immunoglobulin A levels to rule out an immunoglobulin A deficiency and the possible presence of antibodies to IgA. Serum IgG levels in patients were 6·3–13·5 mg/ml, mean 10·2 mg/ml. Serum IgA levels ranged from 0·7 to 3·83 mg/ml, mean 2·6 mg/ml and IgM levels from 0·76 to 3·7 mg/ml, mean 2·0 mg/ml.

Tests for the presence of rheumatoid factor by Waaler Rose agglutination and latex fixation were negative in all but one patient, in whom the titres were 50 and 200 IU, respectively.

Studies in healthy controls and in patients with SLE

Based on a previous study (Lobatto *et al.*, 1987), a dose of 10 μg ^{123}I-AIgG/kg body weight was chosen for this study. Nine healthy controls and 15 patients with SLE, respectively received a mean of 680±85 (s.d.) and 610±63 μg radiolabelled AIgG intravenously as a 3 ml bolus in 30 s through a plastic cannula in the forearm.

Radioactivity detectable over the hepatic, splenic, and cardiac regions was registered continuously during 1 h. Serial blood samples were obtained from the opposite forearm through a plastic cannula during 1 h to determine residual radioactivity in whole blood and plasma. Protein bound radioactivity was measured by precipitating the samples with 10% trichloroacetic acid. Since the ^{123}I-AIgG preparation also contained approximately 40% monomeric AIgG, the clearance of polymeric AIgG was assessed by measuring radioactivity of precipitates obtained by treating plasma samples with polyethyleneglycol 6000 at a final concentration of 3% at 0°C. The AIgG preparation used in this study has been shown to activate complement (Lobatto *et al.*, 1987). Erythrocyte bound radioactivity was assessed in fractions of 5×10^8 erythrocytes obtained after washing once with 5 ml PBS at 0°C. The degree of complement activation after infusion of AIgG was determined in EDTA-plasma samples, by assessment of C3a-levels by sandwich Elisa (Lobatto *et al.*, 1987).

Immune complex assays

These were performed with the ^{125}I-C1q binding assay (Zubler *et al.*, 1976), and the results were expressed in μg equivalent of aggregated IgG as previously described (Kauffman, Van Es & Daha, 1979).

Determination of the number of CR1 receptors on erythrocytes

Extensively washed erythrocytes (5×10^8) in lightly siliconized glass tubes were incubated with an excess of ^{125}I-anti-CR1 monoclonal antibody 2A8 in a final volume of 100 μl RPMI-BSA at 4°C for 16 h. After washing three times with 1 ml RPMI-BSA at 4°C, the cell-associated ^{125}I-anti-CR1 was determined, using tubes without cells incubated with ^{125}I-anti-CR1 as a reagent blank. Non-specific binding to tubes was usually 0·1%. Assays were performed in duplicate. It was shown using dimeric C3b (Daha & Van Es, 1982) that an average of one molecule of anti-CR1 was bound to one CR1 molecule. To correct for inter-assay variation, a standard of human erythrocytes with a known number CR1 per erythrocyte was included in each assay. The standard erythrocytes were kept in fractions at −70°C, thawed before each assay, and used within 2 days.

Statistical analyses

These were performed with Student's *t*-test for unpaired samples, the Mann-Whitney *U*-test and linear regression analysis. Blood curves were analysed for the disappearance of ^{123}I-AIgG from the circulation using a method described by Van der Woude *et al.* (1986).

RESULTS

Studies in healthy controls and patients with SLE

Nine controls and 15 patients with SLE received a dose of 10 μg ^{123}I-AIgG/kg body weight intravenously. No changes in blood pressure or pulse rate were detected during, or within 1 h after, the infusion of AIgG in any of the participants in the study. No complaints were noted during or after the study.

Radioactivity in whole blood, plasma, and trichloroacetic acid precipitable activity in both patients and volunteers showed an initial and rapid distribution phase and a second, slower elimination phase. Polyethyleneglycol (PEG) precipitable radioactivity showed a similar rapid distribution phase, but the elimination phase had a more rapid rate than trichloroacetic acid precipitable activity. Figure 1 shows an example of this in a patient with SLE. The $t_{\frac{1}{2}}$ for the distribution phase obtained for SLE patients (mean±s.d.: 3·5±2·7) was not significantly different from controls (4·6±1·7). The elimination phase of

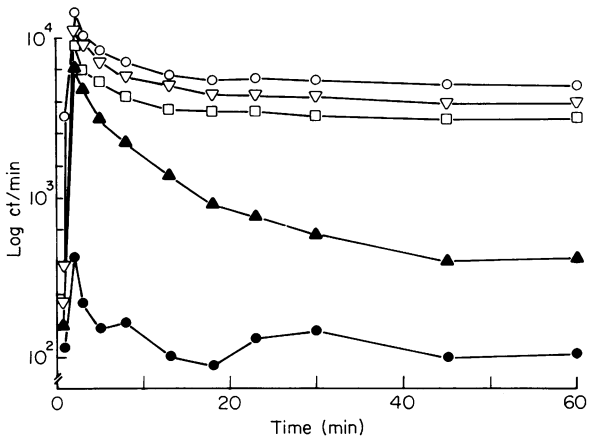

Fig. 1. Radioactivity in blood samples drawn at various times after infusion of ^{123}I-AIgG in a patient with systemic lupus erythematosus. (□) whole blood, (○) plasma, (▽) plasma treated with trichloro-acetic acid, (▲) plasma treated with polyethyleneglycol 6000 at a final concentration of 3%, and (●) erythrocyte bound radioactivity.

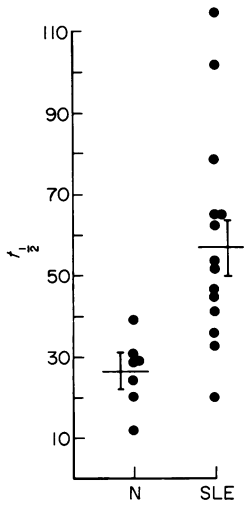

Fig. 2. Elimination rates of ^{123}I-AIgG ($t_{\frac{1}{2}}$) in healthy controls (left) and patients with systemic lupus erythematosus (right). $t_{\frac{1}{2}}$ is significantly longer in patients ($P < 0.005$).

PEG precipitable radioactivity in SLE patients, however, had a significantly longer mean $t_{\frac{1}{2}}$ value of 58 ± 27 min than the $t_{\frac{1}{2}}$ of 26 ± 8 min in controls ($P < 0.005$; see Fig. 2). The ^{123}I-AIgG used in the present study was shown to activate complement *in vitro* (Lobatto *et al.*, 1987), so the degree of complement activation after administration of AIgG in patients and volunteers was measured. No changes in initial levels (Table 1) of CH50, C3, C4, or C1q were observed in either of the two groups. Initial C3a levels were 65 ± 10 mg/ml in the patient group and 115 ± 69 mg/ml in the control group. In the controls C3a levels rose to 207 ± 149 mg/ml following AIgG administration and decreased to baseline levels after 10–15 min. In contrast, the patients showed no clear increase in C3a levels. Binding of ^{123}I-AIgG to erythrocyte CR1 receptors was significantly lower in SLE patients than in healthy volunteers, $9{\cdot}3 \pm 8{\cdot}1\%$ for the patients and $24 \pm 20\%$ in the volunteers ($P < 0{\cdot}05$) at 1 min after infusion of AIgG. At subsequent points in time, the difference was no longer significant. Because the SLE patients as a group had a significantly lower number of CR1 receptors on erythrocytes than healthy controls (Table 1, $P < 0{\cdot}05$), a correlation between the elimination rate and CR1 on erythrocytes was sought, but could not be found. Although there was also a significant diference in CR1 between normo-complementemic and hypo-complementemic patients ($P < 0{\cdot}05$), $t_{\frac{1}{2}}$ did not differ significantly between these two groups. A correlation between percentage of ^{123}I-AIgG-binding, representing binding to CR1 receptors, and C3a-levels was not present either.

Analysis of ^{123}I-AIgG clearance

Since these results suggest defective clearance of ^{123}I-AIgG in SLE, the sites of removal of AIgG from the circulation were analysed. The levels of radioactivity of ^{123}I-AIgG detectable over liver, spleen, and heart (representing blood) as measured with a gamma-camera, are shown in Fig. 3 for a patient with SLE and a control. In this figure, it can be seen that hepatic uptake is higher than splenic uptake in both, but in the SLE patient splenic uptake is minimal. Because the peak level of radioactivity in the liver was reached at $15 \pm 1{\cdot}3$ min in patients and controls, data for hepatic and splenic uptake at 15 min after injection of ^{123}I-AIgG in patients and controls were compared. Although the data obtained for hepatic uptake does not differ significantly for the two groups, splenic uptake is significantly lower in the patients with SLE ($P < 0{\cdot}0005$). Because the results obtained with radioisotope scans differ between individuals depending on geometric and physical factors, it is difficult to compare absolute results between individuals. In order to achieve a degree of standardization, liver/spleen uptake ratios were also determined. These ratios were significantly higher in SLE patients than in controls (Fig. 4) at 15 min, $3{\cdot}9 \pm 2{\cdot}0$ vs $2{\cdot}3 \pm 0{\cdot}7$ ($P < 0{\cdot}05$). Also, ratios were determined at the end of the study, 60 min after injection of AIgG. These were $2{\cdot}4 \pm 1{\cdot}2$ in SLE patients and $1{\cdot}4 \pm 0{\cdot}45$ in volunteers ($P < 0{\cdot}05$).

Relationship between elimination rate of ^{123}I-AIgG and disease activity and laboratory parameters

In the patient group, SLE activity scores ranged from 2 to 20, with a mean of 8·7. In five patients the score was > 10, in another five the score was between five and 10. The remaining five patients had a score < 5. The ages, disease activity scores, disease duration, and medication of patients are shown in Table 2.

The elimination rate ($t_{\frac{1}{2}}$) and organ uptake of ^{123}I-AIgG were analysed in relation to disease activity. No correlation was found between disease activity scores and the elimination rate, or liver/spleen uptake ratios at 15 and 60 min after injection of ^{123}I-AIgG. Separate analyses of patient groups with inactive, moderately active, and highly active disease did not demonstrate a difference in either $t_{\frac{1}{2}}$, liver/spleen uptake ratio, or spleen uptake between the three groups of patients. An effect of medication on $t_{\frac{1}{2}}$ or liver/spleen uptake ratios also could not be established.

To determine a possible relationship between elimination of ^{123}I-AIgG and circulating immune complex levels, $t_{\frac{1}{2}}$ in SLE patients was correlated with circulating immune complex levels as detected by the C1q-binding assay; but no correlation was found between the $t_{\frac{1}{2}}$ and levels of C1q-binding immune complexes.

DISCUSSION

In this study we found that the removal of AIgG from the circulation is abnormal in patients with SLE. The $t_{\frac{1}{2}}$ of AIgG in the circulation was found to be prolonged, and the uptake of AIgG in the spleen was decreased when compared to healthy volunteers.

Our current findings add evidence to the previous findings using E.IgG (Frank *et al.*, 1979; Hamburger *et al.*, 1982) that splenic Fc receptor dysfunction exists in SLE. The results obtained with AIgG in this study may not only reflect an Fc receptor dysfunction, but also a C3b(CR1) receptor dysfunction. Although we found a significant difference in $t_{\frac{1}{2}}$ and liver/spleen uptake ratio between controls and SLE patients, these findings did not correlate with circulating immune complex levels or with disease activity. The lack of correlation between $t_{\frac{1}{2}}$ and disease activity might be due to the fact that not all disease manifestations in SLE are due to the presence of circulating immune complexes or their inefficient clearance. The significantly lower splenic uptake of AIgG was also not correlated with disease activity. If AIgG would be handled as circulating

Table 1. Values of complement and immune complex levels in volunteers and SLE patients and CR1 receptor numbers on erythrocytes

	CH50	C1q (mg%)	C4 (mg%)	C3 (mg%)	C1q-binding assay (μgreq AIgG/ml)	CR1 no/E
Controls ($n=9$)	270±73	12±1	19±4	65±12	0±0	862±72
Normocomplementemic patients ($n=7$)	341±61	14±3	23±7	79±16	7±14	801±95
Hypocomplementemic patients ($n=8$)	99±57	9±4	7±7	57±19	51±56	614±185

AIgG, aggregated human immunoglobulin G; E, erythrocyte; μgreq, microgram equivalent.

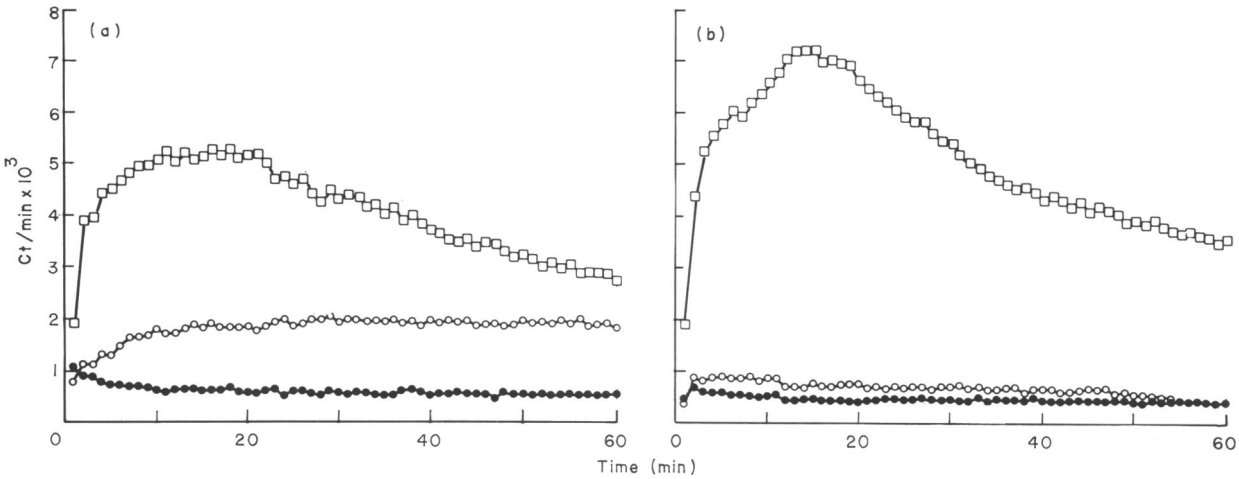

Fig. 3. Liver (□), spleen (○), and heart (●) uptake curves of ^{123}I-AIgG during 1 h after infusion (a) in a healthy control and (b) in a patient with SLE. Splenic uptake is minimal in the patient.

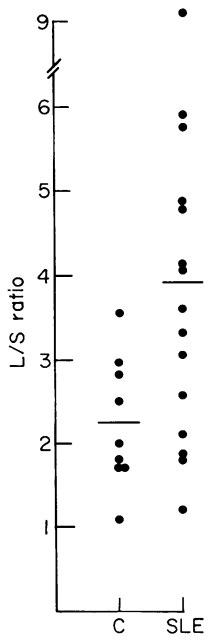

Fig. 4. Liver/spleen uptake ratios in healthy controls and patients with SLE. This ratio is significantly higher in the patient group ($P<0.05$), due to diminished splenic uptake of AIgG.

Table 2. Age, disease duration and medication of SLE patients

	Group I score < 5 ($n=5$)	Group II score 5–10 ($n=5$)	Group III score > 10 ($n=5$)
Age (mean ± s.d.)	47·2(10)	47·2(19·4)	34·7(4·3)
Duration of disease in years	11·8 (3)	6·1 (3·4)	8·2(6·1)
Medication (number of patients using each drug)			
Prednisone			
< 20 mg/day	0	0	2
> 20 mg/day	4	5	3
Azathioprine	1	3	4
Antimalarials	2	2	0
NSAID	2	1	0

NSAID, non-steroidal anti-inflammatory drugs.

immune complexes in SLE, a correlation between the elimination rate of AIgG and circulating immune complex levels could be expected. Several explanations can be given for this lack of correlation. First, AIgG was shown by gel-filtration and sucrose density gradient ultracentrifugation to be composed of aggregates of a wide size range (Lobatto *et al.*, 1987), which might differ in composition from the C1q reactive material in the circulation of SLE patients. It is possible, therefore, that various

correlations may have been obscured, and that the use of a probe of a more homogenous aggregate size would have resulted in other findings. Secondly, the number of patients in our study with substantial levels of circulating immune complexes was small. Another possible explanation is that the $t_{\frac{1}{2}}$ of AIgG reflects mononuclear phagocyte system function, but that the level of circulating immune complexes in SLE patients is not only influenced by their elimination rate, but also by the rate of formation.

A role for erythrocyte C3b receptors in the removal of preformed immune complexes from the circulation has been established in baboons (Cornacoff *et al.*, 1983). We found that erythrocytes of patients with SLE play a role in the removal of AIgG from the circulation, because they were shown to bind AIgG. The magnitude of binding of AIgG to erythrocytes was similar in SLE patients and controls, except at 1 min after infusion, when binding was higher in controls. Because SLE patients have a lower density of C3b receptors on erythrocytes than controls (Iida, Monaghi & Nussenzweig, 1982; Wilson *et al.*, 1982; Walport *et al.*, 1985), less binding of AIgG to erythrocytes would have been expected in patients if saturating doses had been used. However, the dose of AIgG administered in this study can not be expected to saturate all receptor sites on erythrocytes in controls or SLE patients. Therefore, it is not surprising that differences in AIgG binding were not found.

In contrast to a recent report (Belmont *et al.*, 1986), we found that patients with SLE had low basal C3a levels, and failed to show the substantial rise in C3a-levels which we found in healthy volunteers. Several explanations for these findings are possible. If C3a is cleared from the circulation more rapidly in patients with SLE than in controls, this could explain the absence of a rise in C3a-levels in the patients. Another explanation could be that patients with SLE have an increased threshold for complement activation. The mechanism involved needs to be investigated.

The results of this study show that SLE patients eliminate AIgG less efficiently than controls. The relatively low degree of AIgG binding to erythrocytes and the small differences in erythrocyte binding between patients and controls do not fully explain the slower elimination rate in SLE patients.

In conclusion, we have established a dysfunction of the mononuclear phagocyte system in removing AIgG from the circulation in patients with SLE, possibly related to a splenic Fc and C3b receptor abnormality.

ACKNOWLEDGMENTS

The authors wish to thank Dr M.L. Westedt for critical comments regarding the manuscript. Thanks are due to Mrs A.H. Piket-Groenendaal and Mrs H.C.N. Kappelle-de Vries for typing the manuscript. Excellent statistical advice was given by Dr J. Hermans. The technical help provided by Ms R. Bok, Mr J.A.J. Camps and Mr J. Feitsma was greatly appreciated.

REFERENCES

BELMONT, H.M., HOPKINS, P., EDELSON, H.S., KAPLAN, H.B., LUDWIG, R., WEISSMANN, G. & ABRAMSON, S. (1986) Complement activation during systemic lupus erythematosus. *Arthritis Rheum.* **29,** 1085.

CORNACOFF, J.B., HEBERT, L.A., SMEAD, W.L., VANAMAN, M.E., BIRMINGHAM, D.J. & WAXMAN, F.J. (1983) Primate erythrocyte-immune complex-clearing mechanism. *J. clin. Invest.* **71,** 236.

DAHA, M.R. & VAN ES, L.A. (1982) The role of cellular Fc and C3 receptors on the complement-dependent degradation of stable soluble immunoglobulin aggregates by normal and trypsin-treated peritoneal macrophages. *Immunology* **47,** 203.

FRANK, M.M., HAMBURGER, M.I., LAWLEY, T.J., KIMBERLY, R.P. & PLOTZ, P.H. (1979) Defective reticuloendothelial system Fc-receptor function in systemic lupus erythematosus. *N. Engl. J. Med.* **300,** 518.

FRANK, M.M., MODERATOR (1983) Immunoglobulin Fc receptor functional activity in autoimmune diseases. *Ann. intern. Med.* **98,** 206.

HAMBURGER, M.I., LAWLEY, T.J., KIMBERLY, R.P., PLOTZ, P.H. & FRANK, M.M. (1982) A serial study of splenic reticuloendothelial system Fc-receptor functional activity in systemic lupus erythematosus. *Arthritis Rheum.* **25,** 48.

IIDA, K., MONAGHI, R. & NUSSENZWEIG, V. (1982) Complement receptor (CR1) deficiency in erythrocytes from patients with systemic lupus erythematosus. *J. exp. Med.* **155,** 1427.

KAUFFMANN, R.H., VAN ES, L.A. & DAHA, M.R. (1979) Aggregated immunoglobulin G stabilized by albumin: a standard for immune complex detection. *J. immunol. Methods* **31,** 11.

KIMBERLY, R.P., PARRIS, T.M., INMAN, R.D. & MCDOUGAL, J.S. (1983) Dynamics of mononuclear phagocyte system Fc receptor function in systemic lupus erythematosus. Relation to disease activity and circulating immune complexes. *Clin. exp. Immunol.* **51,** 261.

KIMBERLY, R.P. & RALPH, P. (1983) Endocytosis by the mononuclear phagocyte system and autoimmune disease. *Am. J. Med.* **74,** 481.

LOBATTO, S., DAHA, M.R., VOETMAN, A.A., EVERS-SCHOUTEN, J.H., VAN ES, A.A., PAUWELS, E.K.J. & VAN ES, L.A. (1987) Clearance of soluble aggregates of human immunoglobulin G in healthy volunteers and chimpanzees. *Clin. exp. Immunol.* **68,** 133.

MANNIK, M. & AREND, W.P. (1971) Fate of preformed immune complexes in rabbits and rhesus monkeys. *J. exp. Med.* **134,** 19S.

PARRIS, T.M., KIMBERLY, R.P., INMAN, R.D., MCDOUGAL, S., GIBOFSKY A. & CHRISTIAN, C. (1982) Defective Fc receptor-mediated function of the mononuclear phagocyte system in lupus nephritis. *Ann. int. Med.* **97,** 526.

TAN, E.M., COHEN, A.S., FRIES, J.F., MASI, A.T., MCSHANE, D.J., ROTHFIELD, N.F., GREEN SCHELLER, J., TALAL, N. & WINCHESTER, R.J. (1982) The 1982 revised criteria for the classification of systemic lupus erythematosus. *Arthritis Rheum,* **25,** 1271.

VALENTIJN, R.M., VAN OVERHAGEN, H., HAZEVOET, H.M., HERMANS, J., CATS, A., DAHA, M.R. & VAN ES, L.A. (1985) The value of complement and immune complex determinations in monitoring disease activity in patients with systemic lupus erythematosus. *Arthritis Rheum.* **28,** 904.

VAN DER WOUDE, F.J., VAN DER GIESSEN, M., KALLENBERG, C.G.M., OUWEHAND, W., BEEKHUIS, H., BEELEN, J.M., VAN SON, W.J., HOEDEMAEKER, PH. J., VAN DER HEM, G.K. & THE, T.H. (1984) Reticuloendothelial Fc receptor function in SLE patients. I. Primary HLA linked defect or acquired dysfunction secondary to disease activity? *Clin. exp. Immunol.* **55,** 473.

VAN DER WOUDE, F.J., BEEKHUIS, H., BEELEN, J.M. & THE, T.H. (1986) Kinetic analysis of the blood clearance and organ uptake curves of IgG-coated red cells in HLA-typed controls and patients with Wegener's granulomatosis. *Eur. J. Nucl. Med.* **12,** 130.

VEERHUIS, R., KROL, M.C., VAN ES, L.A. & DAHA, M.R. (1986) Difference in clearance kinetics of particulate immune complexes and soluble aggregates of IgG *in vivo*. *Clin. Immunol. Immunopathol.* **41,** 379.

WALPORT. M.J., ROSS, G.D., MACKWORTH-YOUNG, C., WATSON, J.V., HOGG, N. & LACHMANN, P.J. (1985) Family studies of erythrocyte complement receptor type 1 levels: reduced levels in patients with SLE are acquired, not inherited. *Clin. exp. Immunol.* **59,** 547.

WILSON, J.G., WONG, N.W., SCHUR, P.H. & FEARON, D.T. (1982) Mode of inheritance of decreased C3b receptors on erythrocytes of patients with systemic lupus erythematosus. *N. Engl. Med.* **307,** 981.

ZUBLER, R.H., LANGE, G., LAMBERT, P.H. & MEISCHER, P.A. (1976) Detection of immune complexes in untreated sera by a modified ^{125}I-C1q binding test. *J. Immunol.* **116,** 232.