Clin. exp. Immunol. (1987) **69**, 188–197

Fast liver catabolism of C1q in patients with paraproteinaemia and depletion of the classical pathway of complement

J. A. SCHIFFERLI, M. PASCUAL, GERTRAUD STEIGER, M. SCHAPIRA*, J.-E. RYSER†, J. ESTREICHER‡, & A. DASH *Clinique Médicale, * Division de Rhumatologie, † Division de Médecine Nucléaire, and ‡ Division Informatique, Hôpital Cantonal Universitaire, Geneva, Switzerland*

(Accepted for publication 20 January 1987)

SUMMARY

The main clinical features in four patients with IgG_1k paraproteinaemia and acquired complement deficiency included xanthomatous skin lesions (in three), panniculitis (in three) and hepatitis (in two). Hypocomplementaemia concerned the early classical pathway components – in particular C1q. Metabolic studies employing ^{125}I-C1q revealed a much faster catabolism of this protein in the four patients than in five normal controls and three patients with cryoglobulinaemia (mean fractional catabolic rates respectively: 23·35%/h; 1·44%/h; 5·84%/h). Various experiments were designed to characterize the mechanism of the hypocomplementaemia: the patients' serum, purified paraprotein, blood cells, bone marrow cells, or xanthomatous skin lesions did not produce significant complement activation or C1q binding. When three of the patients (two with panniculitis and hepatitis) were injected with ^{123}I-C1q, sequential gamma-camera imaging demonstrated rapid accumulation of the radionuclide in the liver, suggesting that complement activation takes place in the liver where it could produce damage.

Keywords paraproteinaemia cryoglobulinaemia C1q metabolism complement depletion

INTRODUCTION

Acquired complement deficiency of early classical pathway components has been described in a number of different situations. When not related to an SLE-like syndrome (McDuffie *et al.*, 1973; Marder *et al.*, 1978) it has been associated most often with lymphoproliferative disorders of B cell origin, with cryoglobulinaemia or with paraproteinaemia (Gelfand *et al.*, 1979). Such patients have a characteristically low concentration of C1q. Since angioedema attacks related to reduced C1 inhibitor levels are often the main clinical feature of the disease, these patients have been reported to have 'acquired C1 inhibitor deficiency'. However other presentations have been reported: cold induced urticaria, plane xanthomatosis and meningoccocaemia (Costanzi, Coltman & Donaldson, 1969; Jordon *et al.*, 1974; Kövari *et al.*, 1981; Schifferli *et al.*, 1984). In these patients the complement depletion is sometimes less severe, i.e. despite low C1q and C4 concentrations C1 inhibitor level is normal.

Various mechanisms have been shown to be responsible for complement depletion: activation by tumor cells (Schreiber *et al.*, 1976; Hauptmann *et al.*, 1979), by monoclonal paraprotein (Day *et*

Correspondence: Dr J. A. Schifferli, Clinique Médicale, Hôpital Cantonal Universitaire, 1211 Geneva 4, Switzerland.

al., 1976), or by antiidiotypic antibodies directed at the paraprotein (Geha *et al.*, 1985). However the site of complement activation remains undefined in many patients, particularly in those with paraproteinaemia and plane xanthomatosis.

Of the four patients with paraproteinaemia (PARA patients) and complement depletion reported here, three had plane xanthomatosis, two had a peculiar syndrome of panniculitis and hepatitis and another had one episode of panniculitis (M. Pascual, unpublished). Various experiments were performed to define the site of complement activation and the results obtained were compared to similar studies done in three patients with cryoglobulinaemia (CRYO patients).

MATERIALS AND METHODS

Normal IgG, paraprotein and cryoglobulin purification. The IgG fractions containing the IgG paraproteins and normal IgG were purified from fresh sera by Protein-A Sepharose (Pharmacia) followed by gel filtration on Sephacryl S-300 (Pharmacia) in veronal buffered saline (CFD, Oxoid). The monomeric IgG was concentrated to approximately 10 mg/ml using Aquacide II A (Calbiochem). To remove residual aggregated material the preparations were ultracentrifuged at 120000 ***g*** for 60 min and only the upper half of the solutions were used. The cryoglobulins of the three patients described in Table 1 were obtained from serum collected at 37°C and left 24 h at 4°C. The cryoprecipitate was separated by centrifugation at 3000 ***g*** for 10 min followed by three washes in CFD. After incubation for 1 h at 37°C at half the initial volume of serum, the pellet was removed by centrifugation at 3000 ***g*** at 37°C. These soluble cryoglobulins were used directly for the *in vitro* complement activation studies. The IgG subclass, and light chains (kappa and lambda) of the different paraproteins were determined by double immunodiffusion using monospecific antisera (respectively: Miles and Behringwerke).

Complement, immune complex, C1q precipitin measurements. All functional complement assays were done on fresh serum and for cryoglobulinaemic sera these assays were done immediately keeping the sera at 37°C. Haemolytic C4, C1q, CH50 and alternative pathway 50 (AP50) were measured according to described methods (Lachmann & Hobart, 1976; Praz, Barriera & Lesavre, 1982). Antigenic C1r, C1s, C1 inhibitor, C4, C2, C3, and C4 binding protein were measured by single radial immunodiffusion using monospecific antisera (Miles). C1q concentration was determined using purified C1q as standard (Ziccardi & Cooper, 1977). C3d was measured as described by Perrin, Lambert & Miescher (1975) and results expressed in percentage of normal pooled sera activated at 37°C for 60 min by inulin (2 mg/ml). Immune complexes were measured by a C1q binding test (Zubler *et al.*, 1976), and two different assays were used to detect C1q precipitins in serum or IgG fractions of the four PARA patients: gel diffusion against purified C1q (Agnello, Winchester & Kunkel, 1970) and a solid phase C1q assay (Marder *et al.*, 1984). In both assays the positive control was heat-aggregated normal IgG (63°C for 30 min).

In vitro complement activation. This was measured after mixing the test reagent (sera, paraproteins, cryoglobulins) with fresh normal human serum (NHS): (a) pathological sera were mixed at a ratio of 1/1 with normal serum (100 μl/100 μl); (b) paraproteins and cryoglobulins (1 mg) were mixed with fresh serum (100 μl) and adjusted to a final volume of 500 μl with CFD. After incubation at 37°C for 15 min aliquots were removed and the following assays were performed: haemolytic C4, C1q, CH50 and C3 conversion. In similar experiments C3a, C4a, C5a generated in NHS by the purified paraproteins and cryoglobulins were determined using radioimmunoassay kits provided by Upjohn Diagnostics (Kalamazoo).

C1q. This was purified from the serum of one healthy donor (Tenner, Lesavre & Cooper, 1981). Purity was assessed by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) in the presence and in the absence of reducing agents (Laemmli, 1970). No contaminant was detectable by double immunodiffusion and immunoelectrophoresis using anti-whole human and anti-C1q antisera. The specific activity was 100% compared to C1q from the plasma used for its purification. Radiolabelling was done with ^{125}iodine by the lactoperoxidase method using no more than 0·5 μC for 1 μg of C1q (Enzymobead reagent, BioRad). Free iodine was removed by gel filtration (PD10, Pharmacia) in the presence of 1% human serum albumin (HSA, Sigma). The

haemolytic activity of C1q was unchanged by the labelling procedure. SDS-PAGE followed by autoradiography revealed that the ^{125}I iodine bound mainly to the C chain of C1q. The functional integrity of ^{125}I-C1q was further demonstrated by its incorporation into macromolecular C1 (see below) and by more than 90% binding to aggregated IgG in the C1q binding assay. After microfiltration in the presence of 1% HSA, aliquots were stored at −160°C until use. After the radiolabelling procedure all the manipulations were done using sterile buffers and equipment. Sterility testing was done by standard bacteriological methods; the preparation was pyrogen free when tested in two rabbits. ^{123}I iodine was obtained from EIR (Würenlingen, Switzerland). The labelling of C1q with ^{123}I iodine was identical to ^{125}I iodine labelling with respect to the degree of substitution of molecule iodine/molecule C1q $< 1/10$; the ^{123}I-C1q preserved full haemolytic activity, similar incorporation into C1 and binding to aggregated IgG than ^{125}I-C1q.

Incorporation of ^{125}I-C1q and ^{123}I-C1q into C1 in serum was tested by adding the C1q to 100 μl of fresh serum in the presence or absence of 10 mM EDTA, left at room temperature for 15 min, then ultracentrifuged on sucrose gradients (Schifferli *et al.*, 1985b): C1q was 11 S in EDTA-serum and 16 S in serum, i.e., incorporated into C1.

Human metabolic studies. These were approved by the ethical committee. Normal volunteers and patients were injected with 2–5 μC ^{125}I-C1q or 100–200 μC ^{123}I-C1q. Two millilitres of potassium iodide 5% were given twice daily from the day before the start of the study until its end. Blood samples were collected on EDTA at various time intervals. After centrifugation the protein bound and free radioactivity were measured in plasma (determined after precipitation with an equal volume of 20% tricholoro-acetic acid, TCA); pelleted blood cells (erythrocytes and leucocytes) were washed and counted as well. Urine collections were done for 5 days in normal controls and for shorter periods of time in patients (48 or 72 h; collections were stopped when more than 80% of the injected C1q had been excreted in the urine). Results were analysed using a four-compartments model (1:intravascular and 2:extravascular compartments for ^{125}I-C1q, 3:intravascular and 4:urinary compartment for non-protein-bound ^{125}I). The model was expressed in terms of differential equations with unknown parameters to be estimated (mainly the exchange rates between compartments). Calculations were run using a program of solvers of differential equations and optimization methods (Estreicher, Revillard & Scherrer, 1979). Whereas the volumes of the compartments were parameters directly computed by the optimization methods, the extravascular/intravascular (EV/IV) ratios and the fractional catabolic rates (FCR) were deduced from the fitted values of the exchange rates.

Gamma-camera imaging was performed in one control and three patients after the injection of ^{123}I-C1q: whole body scanning was performed at different time intervals to detect any site of radioactivity accumulation, special attention being given to liver, spleen, bone marrow and the skin lesions. ^{123}I accumulation in the liver (and in the heart) was measured from sequential radioactivity counts on antero-posterior images of liver, heart and two background areas. After subtraction of the average background value, specific accumulation was calculated as the ratio of liver counts/blood counts at different time intervals after injection (radioactivity index). Blood counts were calculated by multiplying the counts measured over the heart area in the first image (acquired within 3 min of injection in all cases) by the fraction of initial radioactivity remaining in the blood samples at different time intervals.

In the four PARA patients peripheral blood mononuclear cells and bone marrow mononuclear cells obtained 30 min after the intra-venous injection of ^{125}I-C1q were purified by centrifugation with Ficoll-Hypaque (Pharmacia) and washed six times in Hanks Borate buffered saline (BBS) (Gibco Ltd.). Peripheral blood mononuclear cells from the normal controls studied simultaneously were prepared similarly. The cells were resuspended in the same buffer and used immediately in the different assays described below. One drop of the cell suspensions were put on a slide, dried for 2 h at room temperature and stored at −30°C in a closed plastic bag until tested. Different assays were done with the cell suspensions:

(a) *in vitro* complement activation; the mixture contained: (I) either 100 μl Hanks buffer only, or 2×10^5, or 2×10^6 cells in 100 μl BBS, and (II) either 100 μl of the patients own serum, or 100 μl of purified IgG paraprotein of the patient (10 mg/ml in VBS), or 100 μl of normal IgG (10 mg/ml in VBS), or 100 μl of VBS, and (III) 100 μl normal human serum. After incubation for 60 min at 37°C,

the mixtures were centrifuged and haemolytic C1q, C4 and CH50 measured. (b) ^{125}I-C1q binding to the cell suspensions; mixtures contained: (I)+(II) (both as under (a))+(III) 100 μl ^{125}I-C1q in 1% HSA. The mixtures were incubated 30 min at 37°C or 120 min at 4°C, centrifuged and the percentage binding to the cells in the pellets determined. The protein bound radioactivity of the supernatants of the experiment carried out at 37°C was determined after precipitation with TCA.

Immunofluorescence. Direct and indirect studies were done on 4 μm cryostat sections of frozen unfixed lesional skin biopsies obtained from two patients with plane xanthomatosis using fluorescein isothiocyanate conjugated (FITC) anti-human IgM, IgG, IgA, kappa and lambda light chains, C4 and C3 and rhodamine conjugated anti-human C1q (Berhingwerke) (Schifferli *et al.*, 1985a). The slides with mononuclear cells from the peripheral blood of normal controls and patients, and from the bone marrow of the patients were studied by direct immunofluorescence after fixation in 5% acetic acid+96% ethanol at −20°C for 15 min.

RESULTS

Patients. The main clinical and biological features of the four PARA patients with C1q consumption are presented in Table 1. The three patients with plane xanthomatosis had evidence of an abnormal plasma cell population in their bone marrow. In the same three patients no auto- or hetero-lymphocytotoxic antibodies could be detected. The severity of complement depletion was different from one patient to another, but all had marked alterations of the early classical pathway components and evidence for complement activation *in vivo* (increased C3d). Further, in one patient (M.), the transfusion of fresh frozen plasma (800 ml) did not restore haemolytic C4 concentration to the expected value (expected 25%, measured 5%), and the rise in C4 was short-lived (<0·1% 1 h after the end of the transfusion). To analyse the characteristics of these patients, they were compared to three CRYO patients who had comparable complement depletion (Table 1). To note is that the cryoglobulinaemic sera contained C1q binding material, whereas the sera of the four PARA patients were negative in the C1q binding assay and in two C1q precipitin assays.

In vitro *complement activation.* The three sera of PARA patients with plane xanthomatosis did not produce complement consumption when mixed with NHS. The other sera (patient B. and CRYO) produced significant consumption, i.e. >20% drop in haemolytic C4 and/or >20% C3 conversion. Similar results were obtained when purified paraproteins and cryoglobulins were incubated in NHS: only cryoglobulins and the paraprotein of patient B consumed CH50, haemolytic C4 and/or produced C3 conversion (>20% lower than the values obtained with normal polyclonal IgG). Haemolytic C1q remained unchanged in all. C3a, C4a and C5a generation measured in identical experiments (Table 2) established more precisely that the complement activating capacity was near maximal for cryoglobulins A. and N. (maximal C4 and C3 activation would produce approximately 20 μg/ml of C4a and 50 μg/ml of C3a), intermediate for cryoglobulin L. and paraprotein B. and low for the three other paraproteins. Thus *in vitro* experiments of complement consumption corresponded to *in vivo* complement depletion in CRYO but not in PARA patients.

In vivo *metabolism of C1q.* See Table 3. The fractional catabolic rate (FCR) of C1q was increased in all seven patients studied; compared to the normal controls the FCR was approximately four times faster in CRYO patients and 14 times faster in those with paraproteinaemia. Such very large differences were also observed in C1q distribution with a predominant extravascular compartment mainly in the PARA patients. In all patients the reduced C1q concentration in plasma could be explained by an accelerated catabolism since the calculated synthetic rates were either in or above the normal range. In addition rapid C1q catabolism was also demonstrated by the increase in free ^{125}I (not precipitated by 10% TCA) in plasma starting 1 h after the intravenous injection of ^{125}I-C1q. Further studies were done to localize the site of C1q uptake and catabolism. Gamma-camera imaging was done in one control and in three PARA patients after the injection of ^{123}I-C1q (Fig. 1). In the normal control the radioactivity at 3 min and at 33 min remained mainly in the vascular compartment (91% of the total radioactivity was still in the circulating pool after 2 h), and compared to other tissues, the liver was the site of some radionuclide

Table 1. Patients

Patients	age/sex	Clinical features	Paraprotein (cryoglobulin)	IgG	IgA	IgM (g/L)	Bone marrow		Complement measurements (% NHP)†								C3d (%)‡	C1q binding activity (%)§
							Aspirate	Immuno-fluorescence*	CH50	C1q	C1r	C1s	C1In	C4	C2	C3		
1 B.	59/M	Panniculitis–hepatitis	IgG1 k	11·9	0·6	0·7	10–15% plasma cells	70% k +30% l	10	10	10	33	13	5	31	165	25	3·5
2 C.	48/M	Pl.Xanthomatosis Panniculitis–hepatitis	IgG1 k	35·4	0·5	0·2	10% plasma cells	>90% k	0	13	20	28	17	<1	25	93	22	3·6
3 W.	58/M	Pl.Xanthomatosis Meningoccocemia Panniculitis	IgG1 k	21·2	0·2	0·9	15–20% plasma cells	>90% k	23	7·5	100	93	118	61	50	59	20	2·9
4 M.	58/M	Pl.Xanthomatosis Xanthogranuloma	IgG1 k	17·9	1·1	0·3	15–20% abnormal plasma cells	>90% k	0	15	10	33	11	<1	25	83	20	4·7
5 A.	50/F	Cryoglobulinaemia chronic hepatitis	IgM k‖ IgG kl	14·0	0·7	5·4	50% lympho plasmatoid cells	ND	15	30	25	42	100	16	62	110	17	82
6 N.	51/F	Cryoglobulinaemia glomerulonephritis	IgM k‖ IgG kl	4·9	1·2	5·5	ND	ND	12	38	12	53	80	1	46	78	38	28
7 L.	39/F	Cryoglobulinaemia	IG1 l‖	15·0	ND	ND	10% plasma	ND	19	53	43	57	100	5	42	100	13	87

* Percentage of the plasma cells staining for kappa(k) and lambda (l) light chains.

† NHP, normal human serum pool from 25 controls; normal range: CH50, 75–125; C1q, 54–146; C1r not done; C1s and C1 In, 60–140; C4, 48–152; C2, 60–140; C3, 68–132. Lytic alternative pathway and C4 binding protein measurements were normal in all patients.

‡ C3d in normals is <5%.

§ C1q binding activity in normals is <10%.

‖ Cryoglobulin concentrations: A, 6·1 mg/ml; N., 3·7 mg/ml; L., 1 mg/ml, patient on weekly plasma exchange which were stopped for 2 weeks before the present investigations.

ND, not done.

Table 2. Complement activation by purified IgG fractions and cryoglobulins*

IgG or cryoglobulin	C4a (μg/ml)	C3a (μg/ml)	C5a (ng/ml)
1 IgG B.	5·31	17·9	182
2 IgG C.	3·30	14·4	166
3 IgG W.	1·56	13·9	141
4 IgG M.	2·58	12·9	141
5 Cryo. A.	13·71	24·9	288
6 Cryo. N.	19·17	25·8	414
7 Cryo. L.	4·00	15·7	116
Normal IgG	1·26	11·1	92
Buffer	0·87	10·8	62

* Incubated 15 min at 37°C with fresh human serum.

Table 3. ^{125}I-C1q metabolic studies

Patients	C1q (μg/ml)	Plasma volume (ml/kg)	EV/IV ratio*	FCR† (%/h)	Synthetic rate (μg/kg/h)
1 B.	7	53·1	4·58	35·20	130·8
2 C.	9	55·7	3·49	15·46	77·5
3 W.	5·25	52·9‡	8·40	21·24	59·0
		(116·8)	(4·60)	(10·69)	(57·4)
4 M.	10·5	67·1	7·35	21·10	148·7
5 A.	21	67·4	1·14	7·20	101·9
6 N.	26·6	65·6	1·37	6·47	112·9
7 L.	37	42·0	0·30	3·94	61·2
Normal controls (5)§	60·5	53·1	0·73	1·44	45·3
	(49–66·5)	(48·6–62·7)	(0·41–0·99)	(1·25–1·71)	(35·8–62·8)

* EV/IV, extra-vascular/intra-vascular distribution.

† FCR, fractional catabolic rate.

‡ Values calculated by fixing the plasma volume to 52·9 ml/kg; the values obtained directly from the linear model chosen are given in parentheses.

§ Mean values of the five normal controls and range: ( ).

accumulation shortly after injection but liver radioactivity did not increase in subsequent images. In contrast in the three patients ^{123}I-C1q accumulated progressively in the liver, and in the spleen (in patient W. only) (Fig. 1c–h). The liver uptake was determined by a specific radioactivity index (see Materials and Methods) which increased in the patients but not in the control (Fig. 2). ^{123}I-C1q catabolism was apparent in late images: 2 to 3 h after injection part of the radionuclide was present in the bladder and to a small extend in the stomach (not illustrated). Radionuclide accumulation was detected in no other site. To check that fast liver uptake of C1q was not due to reduced incorporation of C1q into the macromolecular C1 complex, the serum samples obtained 7·5 min after the injection of ^{123}I-C1q were analysed by sucrose gradient density ultracentrifugation: in all instances (one control and three patients) C1q was completely incorporated into C1 (Fig. 3).

Complement binding and activation by circulating and bone marrow cells, and foamy macrophages of the xanthomatous skin lesions in PARA patients. Immunofluorescence studies were performed on the bone marrow aspirate and the mononuclear cell fraction of the bone marrow. Plasma cells were

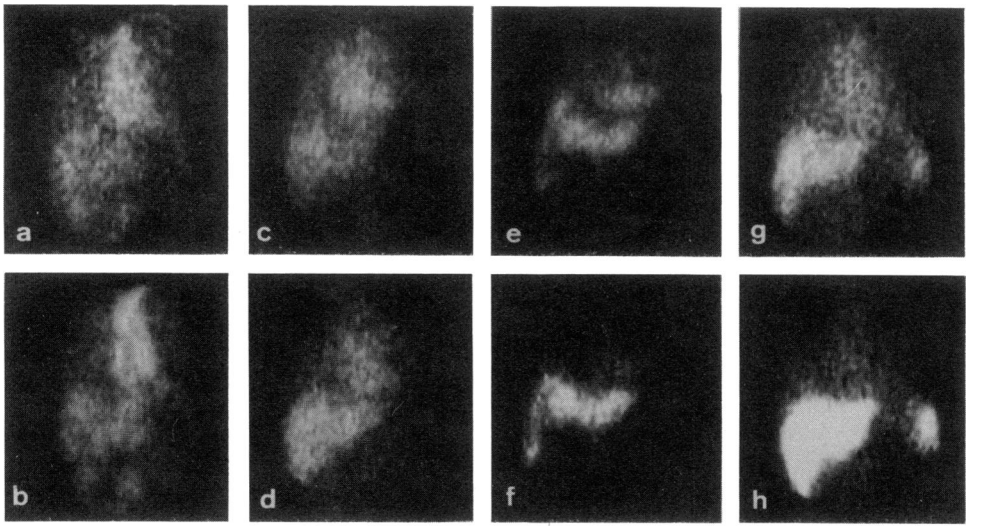

Fig. 1. Gamma-camera imaging made within 3 min (upper row) and between 20 and 33 min (lower row) after the injection of ^{123}I-C1q in a normal control (a, b), and in three patients (C. (c, d); B. (e, f); W. (g, h)): antero-posterior image of the thorax and upper abdomen showing heart + large thoracic vessels, and liver.

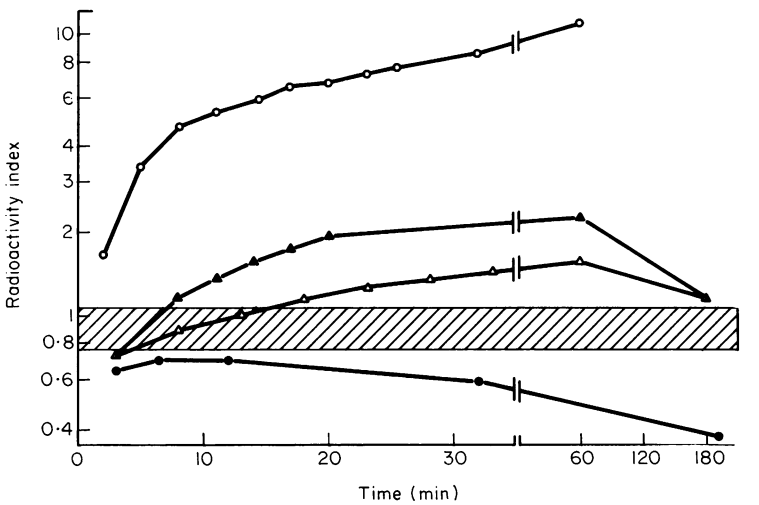

Fig. 2. Time course of the accumulation of ^{123}I-C1q in the liver in three patients and one control (patient B. (▲); C. (△); W. (○); control (●). The liver uptake was determined by the specific liver radioactivity index. The hatched area indicates the control heart radioactivity index which remained constant in all studies.

identified by staining for IgG and kappa light chains, but no C1q, C4 or C3 deposits were detected.

During the ^{125}I-C1q metabolic studies no radioactivity was bound to circulating cells. The bone marrow aspirate was performed 30 min after the i.v. ^{125}C1q injection; a fraction of this aspirate was centrifuged at 3000 *g* for 15 min so as to separate the cellular elements. No radioactivity was found in the pelleted bone marrow cells.

Mononuclear cells of the peripheral blood and bone marrow were purified and incubated with ^{125}I-C1q. Various conditions were used to determine C1q binding and metabolism by these cells, including the addition of the patient's own serum or paraprotein (see Methods). Five to ten percent

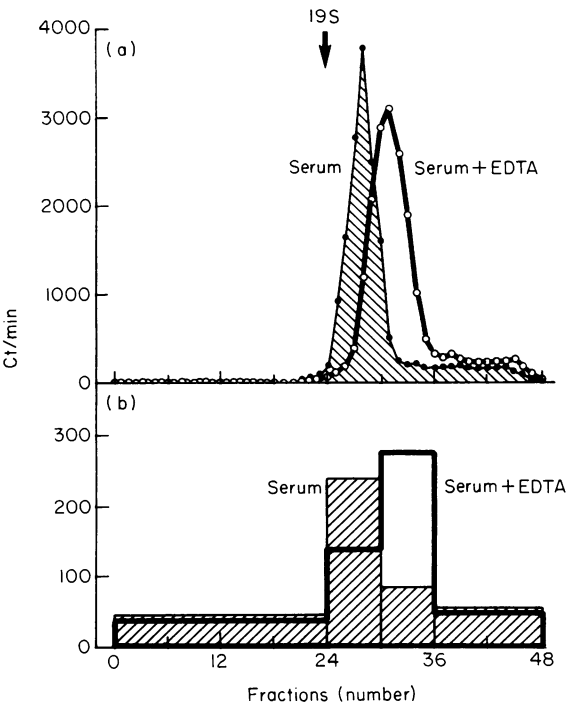

Fig. 3. ^{123}I-C1q incorporation into macromolecular C1 in serum of patient B. was analysed by sucrose density gradient ultracentrifugation. (a) *in vitro*: ^{123}I-C1q was incorporated into C1 in serum and dissociated from C1r-s by the addition of 10 mM EDTA (48 fractions were collected). (b) *in vivo*: similar incorporation into C1 was detected in serum taken 7·5 min after injection of ^{123}I-C1q (since very little radioactivity was present in the serum sample only four unequal fractions were collected). Ct/min, counts per minute. Identical results were observed in the two other patients and the control.

C1q binding to the pelleted cells was found under all conditions including controls, so there was no specific C1q binding. This absence of binding was not due to rapid catabolism since ^{125}I remained protein bound in all supernatants.

Similarly experiments were performed to see if the patients' peripheral blood or bone marrow mononuclear cells mixed with normal serum activated complement *in vitro*. There was no significant loss of haemolytic C1q, C4 and CH50 due to the presence of the cells. Adding patient's own serum or paraprotein to the mixture did not modify the results except in the set of experiments in which serum or paraprotein of patient B. were used (significant loss of haemolytic C4 and reduction of CH50 which were similar in the control experiments done without cells). In two patients with plane xanthomatosis (M. and C.), a skin biopsy revealed infiltration with foamy macrophages. No immunoglobulins or complement proteins (C1q, C4, C3) could be detected in these lesions by direct and indirect (preincubation of the sections with NHS for 15 min at 37°C) immunofluorescence.

DISCUSSION

The mechanisms responsible for depleting complement in the patients studied here could be summarized as follow: in the four patients with paraproteinaemia, complement was consumed nearly exclusively in extravascular compartments, mostly liver, and not directly by the paraprotein. In contrast in patients with cryoglobulinaemia, complement depletion was related to the presence of the cryoglobulins. These conclusions are supported by the following observations:

(1) Early classical pathway components were depleted, and C3d was increased in all patients. However the two groups of patients differed. In the CRYO patients C1q depletion correlated with the presence of C1q binding material and *in vitro* complement activation by the cryoglobulins. In contrast the even more severe C1q depletion in PARA patients could not be explained by reactions involving only the paraproteins or serum factors.

(2) The catabolism of radiolabelled C1q was accelerated in CRYO patients, corresponded to the hypocomplementaemia and was compatible with intra- and extra vascular complement activation by the cryoglobulins. In the PARA patients C1q catabolism was extremely fast, greatly exceeding the catabolism observed in the CRYO patients; further, C1q escaped almost immediately from the circulation. These observations together with low or no significant *in vitro* complement consumption suggested that C1q was fixed in a site in very close contact with the blood stream and was causing local complement activation. Indeed some specific binding site for C1q must have been present since C1q is a large molecule unlikely to reach the extravascular compartment by simple diffusion as rapidly as the present data would indicate. However the fast disappearance was not due to C1q remaining free, i.e. in a form capable of reacting with specific receptors for C1q (Tenner & Cooper, 1980).

(3) Since the disappearance and degradation of C1q was so rapid *in vivo,* we were able to demonstrate the site of C1q catabolism using ^{123}iodine radiolabelled C1q and measuring the radioactivity by gamma camera imaging. These studies were done in three patients and they showed that the liver (and spleen in one patient) was the main site of specific C1q uptake and catabolism.

(4) These results indicating C1q catabolism in the liver were consistent with all the negative investigations which had been performed to see whether the tumor cells in the bone marrow, circulating cells or those involved in the xanthomatous skin lesion were directly involved in complement activation and/or C1q binding. Such negative findings suggested that complement activation in the four patients presented here was not related to monoclonal IgG-antiidiotypic antibodies reactions occurring at malignant cells surfaces as observed in other patients (Geha *et al.*, 1985).

Fast C1q catabolism has been observed in different diseases particularly in patients with the hypocomplementaemic urticarial vasculitis syndrome (Schmid *et al.*, 1977; Curd, Ziccardi & Kaplan, 1979) however such nearly immediate disappearance of C1q has been documented only in rare patients with myeloma with reduced C1q concentration (Kohler & Müller-Eberhard, 1972). Indeed low concentrations of C1q without evidence for complement activation has been observed in patients with myeloma (Kohler & Müler-Eberhard, 1969). One of the patients studied here (patient W.) would almost have fitted into this category but developed profound C4 depletion at a later state (M. Pascual, unpublished). The mechanism of C1q depletion in our patients and in patients with myeloma with reduced C1q concentration could be related.

More work is required to understand why the liver was the site of C1q catabolism and, probably, complement activation in these patients. However it could well be that this activation was not without harm since two of the patients presented a unique clinical syndrome characterized in part by hepatitis.

This work was supported by grants from the Fonds National Suisse de la Recherche Scientifique (3.915–0·85) and the Swiss Federal Department of Public Health; J.A.S. is supported by the Max Clöetta Foundation, and M.P. by the Sir Jules Thorn Charitable Overseas Trust. We thank Dr L. Perrin for performing the C3d and the C1q binding assay, Dr M. Jeannet for testing for lymphocytotoxic antibodies, and Upjohn Diagnostics for kindly providing the C3a, C4a and C5a immunoassay kits.

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