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NEPHROTIC SYNDROME, MONOCLONAL GAMMOPATHY AND AUTO-IMMUNE HYPERLIPIDAEMIA

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SUMMARY

Hyperlipidaemia, glomerulonephritis and IgG λ monoclonal gammopathy were found to be associated in two patients (Ler. and Lac.). A severe atherosclerosis was also present in the former patient. After a purification–activation procedure active IgG λ was obtained from the serum chylomicra and from the ammonium sulphate-precipitated serum gamma-globulins. In passive haemagglutination tests, including inhibition tests, the IgG λ Ler. reacted specifically with the protein moiety of human low density lipoproteins (LDL) and the IgG λ Lac. reacted with human, rat and rabbit very low density lipoproteins (VLDL) as well as with human serum albumin (HSA). These IgG λ behaved like autoantibodies though no activity was found in whole serum, in which they are probably blocked by the corresponding antigens. They are very similar, although not identical, to the autoantibodies found in auto-immune hyperlipidaemia (AIH) and it is proposed that in these patients the hyperlipidaemia is a variety of AIH. Finally it is suggested that the hyperlipidaemia associated with glomerulonephritis chiefly in the nephrotic syndrome may sometimes be AIH. If this is the case the renal lesions and the hyperlipidaemia may be different expressions of an immune complex disease which may also lead to other tissue lesions such as atherosclerosis.

INTRODUCTION

Hyperlipidaemia is one of the most striking features of the nephrotic syndrome. In early reports it was suspected that it might be causally related to the deficiency in serum albumin, secondary to a marked proteinuria (Rosenman, Friedman & Byers, 1956). However, the relation between serum triglyceride and albumin levels was found to be rather irregular, and an elevation of low density lipoproteins (LDL) rather than the more usual elevation of very low density lipoproteins (VLDL) was noted in cases of nephrosis, with no large deficiency of serum albumin (Baxter, Goodman & Havel, 1960).

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More recently, the rise in total blood triglycerides and VLDL in nephrotic children was found to show an inverse correlation to the plasma post-heparin lipase activity (Yamada & Matsuda, 1970); and in antiserum-induced nephrosis in the rat the adipose tissue lipoprotein lipase activity was markedly reduced (Gutman & Shafrir, 1963; Yamada and Matsuda, 1970). However, the way in which this lipoprotein lipase deficiency might induce hyperlipoproteinaemias of different types and not only pure or predominant hypertriglyceridaemias was not explained.

Auto-immune hyperlipidaemia (AIH) is due to the binding activity of the antibody site of immunoglobulins (Ig) (Beaumont, 1965, 1970; Beaumont & Beaumont, 1973). It was first demonstrated in IgA κ myelomas. It was also found in myelomas of other Ig types and in hyperlipidaemias not associated with monoclonal gammopathy. In the present concept, AIH may be induced by a variety of antibodies which inhibit various stages of the complex lipolytic process by which the lipid load is removed from the circulating lipoproteins. In fact, antilipoprotein antibodies which block the surface sites necessary for enzyme attack have been found, as well as anti-enzyme antibodies (Beaumont *et al.*, 1970a; Beaumont & Lemort, 1970). Accordingly, hyperlipo-proteinaemia in AIH may show different patterns.

Since immune disorders, with immune complex deposition in the kidneys, are frequently found in nephrotic syndromes (Blainey *et al.*, 1960; Dixon, Feldman & Vasquez, 1961; Dixon, 1966; Ward & Kibukamusoke, 1969); Costanza *et al.*, 1973) it was suspected that the associated hyperlipidaemia might be AIH. The occurrence in two patients of a benign monoclonal gammopathy together with hyperlipidaemia and a nephrotic syndrome provided an opportunity to answer this question. The present study was undertaken to determine if there were AIH antibodies in the sera of these patients and if the circulating monoclonal Ig was involved.

MATERIALS AND METHODS

Patients (Table 1)

Mr Lac., a man born in 1932, was admitted to hospital in 1970 for a nephropathy with proteinuria, microscopic haematuria, hyperlipidaemia and hypocomplementaemia. The lipoprotein pattern was type IV according to Fredrickson, Levy & Lees (1967) classification, with a large amount of pre-beta-lipoprotein (Fig. 1). At that time the renal function was normal and a biopsy showed a 'membranoproliferative glomerulonephritis' with Ig and C_3 deposition in the glomerular capillaries. Since then, he was followed as an out-patient and was readmitted twice, in 1972 and 1973. Despite treatment with corticosteroids, chlorambucil and indometacine, the nephrotic syndrome remained unchanged and chronic renal failure with hypertension developed. Since October 1972 a γ2 IgG λ (Fig. 2) monoclonal gammopathy was repeatedly found with rather low levels of IgA and IgM. There were no signs of plasmacytoma. From October 1972 to November 1973 several blood samples were taken and examined. During this period the total blood cholesterol varied between 320 and 500 mg/100 ml and total triglycerides between 300 and 3000 mg.

During a period of marked hyperlipidaemia, an intravenous infusion of human albumin was made, which resulted in a dramatic but temporary reduction of the hyperlipidaemia.

Mr Ler., a man born in 1926, was admitted to hospital in 1971, for a nephropathy with proteinuria, microscopic haematuria and hyperlipidaemia. The lipoprotein pattern was type II with an elevation of beta-lipoprotein levels (Fig. 1), and a IgG γ2 (Fig. 2) monoclonal

gammopathy. There was no renal insufficiency, no signs of myeloma and a kidney biopsy showed 'focal and segmental glomerulosclerosis'. From 1971 to 1973, during a 2½-year follow-up the glomerulopathy and the monoclonal gammopathy remained unchanged; the hyperlipidaemia varied with a total cholesterol ranging from 295 to 420 mg/100 ml and total triglycerides of 250–400 mg/100 ml. In 1969 this patient developed a severe ischaemic disease with angina pectoris and a myocardial infarction. Intermittent claudication began in 1972, for which an aortoiliac endarteriectomy was performed in 1973. The arterial lesions were typically atherosclerotic.

Spontaneous agglutination

Spontaneous agglutination of serum lipid particles was looked for under the microscope in the creamy layer which collected at the top of the fresh serum samples kept overnight at +4°C (Beaumont & Lorenzelli, 1972).

TABLE 1. Clinical features of two patients with nephrosis and autoimmune hyperlipidaemia

	Patients	
	Lac.	Ler.
Age (at onset)	38	47
Sex	Male	Male
Proteinuria (g/24 hr)	5	1
Serum albumin (g/100 ml)	1·8	3·2
Monoclonal gammopathy	IgG λ	IgG λ
(Age at first discovery)	(40)	(47)
Triglycerides (range) (mg/100 ml)	300–3000	250–400
Cholesterol (range)	320–520	295–420
Hyperlipoproteinaemia type	IV	II
Serum complement	Very low	
Histological diagnosis	Membranoproliferative glomerulo nephritis	Focal and segmental glomerulo sclerosis
Arterial pressure	Raised	Normal
Renal insufficiency	Yes	No
Arterial atherosclerosis with ischaemic disease	No	Severe

Detection of antibody activity

This was looked for in whole serum and in serum fractions by the double diffusion method of Ouchterlony (1964) in agarose gels and by a passive haemagglutination test (HA test). For these tests purified beta-lipoprotein, albumin and heparin were used. The HA test was performed with rabbit or sheep red blood cells coated with the purified antigens by the chromium chloride method already described (Gold & Fudenberg, 1967, modified by

Beaumont *et al.*, 1969). In this method physiological buffered saline is used as a diluent without any added macromolecules. Appropriate positive controls were set up to make sure that the cells were sensitized by the antigen using rabbit antisera to human beta-lipoprotein and to albumin, or to a human monoclonal IgA λ anti-heparin already described (Beaumont & Lemort, 1970).

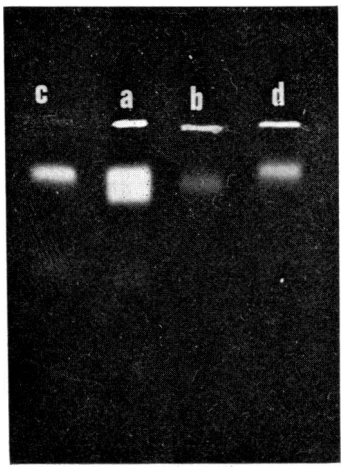

FIG. 1. Lipoprotein patterns after electrophoresis of serum in agarose and staining with Sudan Black. (a) Serum Lac. before and (b) 5 hours after an intravenous albumin infusion. (c) Control serum. (d) Serum Ler.

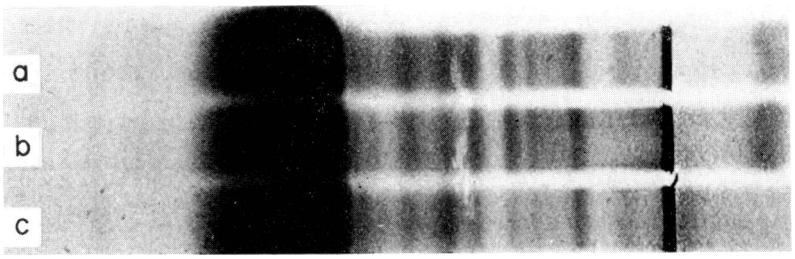

FIG. 2. Protein patterns after electrophoresis of serum in starch gel and staining with Amido Black. (a) Serum Lac. (b) Serum Ler. (c) Control serum.

Specificity of the agglutinating activities

Agglutination of red cells by anti-rabbit or anti-sheep agglutinins was excluded in every case by negative controls with non-coated cells and, if necessary, by absorption of the positive fractions on uncoated cells. Every positive result was controlled by inhibition tests using the following: absorbed whole human serum and serum fractions prepared by gel filtration on Sephadex G-200 and by chromatography on DEAE–cellulose; purified serum lipoproteins; albumin; ovalbumin; heparin; DNA; sugars; phospholipids.

The human serum beta-lipoproteins were prepared either by heparin precipitation and subsequent DEAE–cellulose chromatography according to Poullin, Vilain & Beaumont (1967), which yielded a pool of beta-lipoproteins, or by floatation in the ultracentrifuge according to Havel, Eder & Bragdon (1955), which yielded the following LDL fractions: density <1006, density = 1006–1019 and density = 1019–1063. Both the lipoproteins were carefully washed several times to remove contaminating albumin. Thereafter the 1019–1063 LDL fractions were pure in immunoprecipitation tests. Rabbit and rat serum pooled LDL were prepared in the ultracentrifuge at a density of 1063. Partly delipidated lipoproteins were prepared by extracting purified pooled beta-lipoproteins with a mixture of methanol:ether of 4/96 v:v. The extracted lipids as well as the partly delipidated betalipoproteins were dispersed in saline by sonication and tested in inhibition experiments.

The human serum alpha-lipoproteins were prepared either by DEAE–cellulose chromatography and subsequent gel filtration on Sephadex G-200, according to Beaumont & Lemort (1969), or in the ultracentrifuge at a density of 1·21 after separating the LDL.

Commercial human serum albumin (HSA) and bovine serum albumin (BSA) (Calbiochem.) were used, as well as human serum albumin prepared in the laboratory by DEAE–cellulose chromatography followed by gel filtration on Sephadex G-200.

Activation and purification of the circulating antibodies

From the results obtained by studying auto-immune hyperlipidaemia, it was postulated that the major part, if not all the possible antibody activity would be blocked by a circulating antigen. Several methods were used to dissociate antigen–antibody complexes and subsequently to prepare active antibody. The following method was found to be best. It has two steps. The first step is a preparative ultracentrifuge run in a medium of density = 1·21 containing 0·25% sodium oleate according to a procedure described elsewhere (Beaumont *et al.*, 1970b). In this step the lipoproteins are separated from the antibody by the detergent and move towards the top of the tube. The immunoglobulins, on the other hand, move towards the bottom and are collected with a tube piercer.

The second step is an affinity chromatography in which a CNBr–Sepharose 4-B gel linked with purified beta-lipoprotein or albumin is used as an immunosorbent.

The CNBr-Sepharose beta-lipoprotein or albumin gels are prepared with CNBr-activated Sepharose 4-B (Pharmacia, Uppsala) according to Axen, Porath & Ernback (1967).

It was possible to link 10 mg of albumin and 17 mg of beta-lipoprotein polypeptide to each gram of dry Sepharose. After blocking and washing, the gels were used as follows.

The proteins obtained in the ultracentrifuge run are dialysed overnight against buffered saline at pH 7. They are then dialysed for 2 hr in 0·2 M Tris buffer containing 0·15 M NaCl, at pH 8, and are then incubated for 2 hr in a rotating tube with CNBr–Sepharose beta-lipoprotein or albumin gel equilibrated with the same solvent. The gel is then layered in a column and washed with Tris buffer, 0·15 M NaCl, pH 8, until the OD of the effluent at 280 nm remains at zero. Lastly the proteins retained by the column are eluted with glycine buffer, 0·2 M, pH 3, containing 0·5 M NaCl, and concentrated by dialysis, either by freeze-drying or by negative pressure dialysis.

This procedure was performed on the following serum fractions prepared from the sera of the two patients: chylomicra, prepared by floatation at density = 1006 and at 18,000 r.p.m. for 1 hr; and the washed $(NH_4)_2SO_4$ precipitate obtained at 40% saturation after separating the chylomicra.

The agglutinating proteins

Proteins were measured according to Lowry *et al.* (1951).

The proteins present in the active fractions were detected by agarose double diffusion immunoprecipitation tests with the following antisera (Hyland): anti-human Igγ, μ, α, λ and κ chains; anti-human beta-lipoprotein; alpha-lipoprotein; albumin; anti-whole normal serum.

The agglutinating activity of the active fractions was checked after absorbing with anti-Ig γ, μ, α, λ and κ chains.

RESULTS

Agglutination of circulating lipid particles

After overnight storage at +4°C, there was no notable agglutination. However, a fine agglutination of the chylomicra was seen under the microscope in some of the samples studied.

Whole serum and unprepared serum fractions were inactive

In double diffusion experiments whole serum, chylomicra and the 40% $(NH_4)_2SO_4$-treated fractions did not precipitate with human beta-lipoproteins, HSA or heparin. In HA tests, after appropriate absorption with non-coated RBC, they did not agglutinate RBC coated with beta-lipoproteins, HSA or heparin.

TABLE 2. Results of haemagglutination tests in two patients with autoimmune hyperlipidaemia

Serum and serum fractions tested	Agglutination of RBC sensitized with:	
	Human beta-lipoprotein pool	Human serum albumin
Rabbit anti-human beta-lipoprotein	+++ 1/10,000*	—
Rabbit anti-human serum albumin	—	+++ 1/100,000*
Ler. whole serum	—	—
Lac. whole serum	—	—
Ler. IgG λ purified†	+++ (0·01)‡	—
Lac. IgG λ purified†	+++ (0·5)‡	+++ (0·1)‡

* Highest dilution which yields a +++ agglutination of sensitized RBC.

† By ultracentrifugation in a sodium oleate-containing medium and subsequent affinity chromatography starting from chylomicra or $(NH_4)_2SO_4$-precipitated γ-globulins.

‡ Lowest quantity in micrograms which still gives a +++ agglutination of sensitized RBC.

Fractions prepared by the activation and purification procedure were active in HA tests (Table 2)

In the concentrations available the proteins issued from the chylomicra and the 40% $(NH_4)_2SO_4$ serum fractions after the combined ultracentrifuge oleate and immunosorption procedure did not react in double diffusion experiments, but they did agglutinate the coated RBC (Fig. 3).

For Lac. the most reactive fraction was obtained after immunosorption on Sepharose–

CNBr–HSA columns. It agglutinated both RBC coated with beta-lipoproteins or HSA. However, the titre was higher with HSA-coated RBC, with which 0·1 μg of the protein still gave a + + + agglutination pattern.

For Ler. the reactive fraction was obtained after immunosorption on Sepharose–beta-lipoprotein columns and no activity was obtained with Sepharose–HSA. It agglutinated

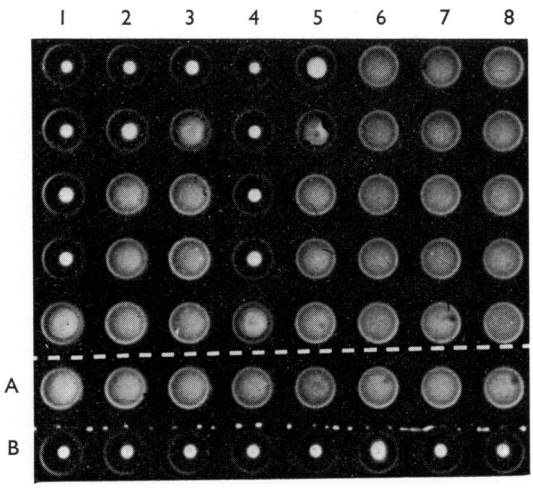

FIG. 3. Passive haemagglutination inhibition test using beta-lipoprotein-sensitized RBC and 1 μg of IgG λ Ler. extracted from the chylomicra by the activation–purification procedure. The tested inhibitors are: (1) beta-lipoprotein obtained by heparin precipitation of pooled human serum; (2) VLDL, density < 1006; (3) LDL, density = 1006–1019; (4) LDL, density = 1019–1063; (5) α-lipoprotein; (6) HSA; (7) BSA; (8) heparin. (A) Positive controls. (B) Negative controls.

TABLE 3. Results of the activation and purification procedure (see Materials and Methods section)

		Proteins prepared		
	Starting serum fractions	Micrograms	Ig type	Other proteins
Serum Lac. (4 ml)	Chylomicra	150	IgG λ	—
	40% $(NH_4)_2SO_4$ precipitate	225	IgG λ	—
Serum Ler. (4 ml)	Chylomicra	150	IgG λ	Albumin traces
	40% $(NH_4)_2SO_4$ precipitate	350	IgG λ	—

only beta-lipoprotein-coated RBC and not HSA-coated RBC. The end point for a + + + pattern was 0·01 μg of protein.

The reactive fractions of Ler. or Lac. did not agglutinate heparin-coated or non-coated RBC. They did not lose their reactivity after absorption on non-coated RBC. No activity was found in similar fractions prepared with normal human sera.

The active proteins were monoclonal IgG

With the exception of traces of albumin in some instances, the only protein found in the active fractions was an IgG λ (Table 3, Fig. 4). The agglutinating activity was absorbed by anti-γ and anti-λ antisera but not by anti-μ, anti-α or anti-κ.

Starting from 4 ml of serum 3–500 μg of active protein was usually prepared with the activation purification procedure. This yield did not permit further study of the Ig reactive site.

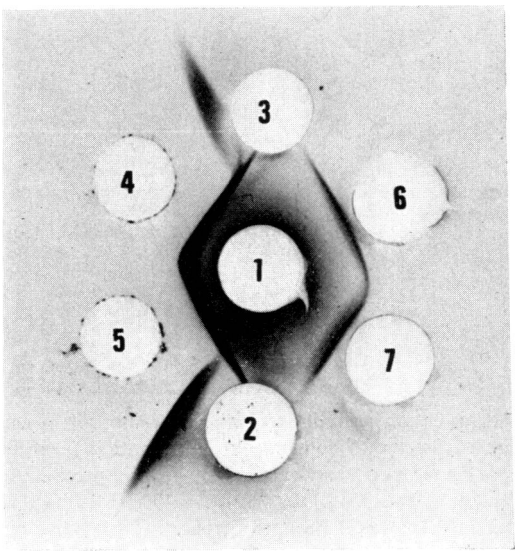

FIG. 4. Identification of the IgG present in active extracts obtained by the activation–purification procedure, starting with the 40% $(NH_4)_2SO_4$ precipitate of serum Ler. Double diffusion experiments: well 1, anti-IgG; well 2, anti-λ chain; well 3, anti-κ chain; well 4 and 5, normal human serum; wells 6 and 7, active extract of Ler.

IgGλ Ler. and IgGλ Lac. reacted with different antigen determinants

As seen in previous cases, IgG λ Ler. and IgG Lac. were both associated with the circulating chylomicra from which they were extracted, as well as from the bulk of the $(NH_4)_2SO_4$ precipitated Ig. However, during the activation and purification procedure IgG Ler. was bound only by the Sepharose–beta-lipoprotein immunosorbent and the eluate reacted only with beta-lipoprotein-coated RBC. On the other hand IgG Lac. was bound by both Sepharose–beta-lipoproteins and Sepharose–HSA immunosorbents and the eluate reacted with either beta-lipoproteins or HSA-coated RBC.

Inhibition experiments showed that the antigen determinants involved in these reactions were different (Table 4). For IgG λ Ler. the antigen determinant was present only in human LDL with a maximum concentration in the 1019–1063 density fraction. Partial delipidation experiments suggest that it is associated with the protein moiety of the molecule.

For IgG λ Lac. the antigen determinant was present: in rat and rabbit as well as in human VLDL—with a maximum concentration in the density <1006 fraction—and in the protein moiety of the lipoprotein molecules as well as in HSA and BSA. As the density <1006

lipoprotein fractions contain some albumin, inhibitions were made with preparations of different purities and with pure HSA. These experiments suggest that the main antigenic determinant is on the HSA molecule, but that the VLDL and chylomicra also interact as inhibitors; perhaps through the albumin molecules which are included in their structure.

IgG λ Ler. and IgG λ Lac. were not inhibited by serum alpha$_1$-lipoproteins, ovalbumin, heparin, DNA and several sugars or phospholipids.

TABLE 4. Antigenic specificity of the two monoclonal antibodies

Molecules tested for inhibition	Inhibition of agglutination* induced by 1 μg of:	
	IgG λ Ler.	IgG λ Lac.
Serum beta-lipoprotein		
Human pool†	+ (0·05)**	+ (1·1)
Human d < 1006‡	+ (5)	+ (0·4)
Human d = 1006–1019‡	+ (4)	–
Human d = 1019–1063‡	+ (0·02)	+ (44)
Human pool partly delipidated	+ (3)	+ (1·2)
Human pool lipids	–	–
Rabbit pool (3)	–	+ (8)
Rat pool (3)	–	+ (3·2)
Serum α lipoprotein human‡	–	
Serum albumin (human)	–	+ (0·02)
Serum albumin (bovine)	–	+ (8)
Ovalbumin	–	–
Heparin	–	–
DNA	–	–
Sugars§	–	–
Phospholipids¶	–	–

* RBC were sensitized with HSA for testing IgG Lac and with human beta-lipoprotein pool for testing IgG Ler.

† Purified by chromatography after heparin precipitation.

‡ Purified by flotation in the ultracentrifuge.

§ Glucuronic acid, D-glucosamine, D-glucose.

¶ Lecithin, sphingomyelin, cephalin, lysolecithin.

** Least quantity which gives complete inhibitions (μg).

Effects of an intravenous HSA infusion on IgG λ Lac.

Within a few hours after this infusion, the serum, which was highly lipaemic and milky, became clear, and its triglyceride content was markedly reduced. An active IgG was extracted from the 40% $(NH_4)_2SO_4$ precipitate of serum taken after the infusion. However, the yield of the activation purification procedure was lower than that of serum taken before the infusion.

DISCUSSION

Although all the immunochemical criteria were not checked, it is likely that IgG λ Lac. and IgG λ Ler. behave as antibodies. They are agglutinating and so at least divalent, and they

react with specific structures. Furthermore, this specificity is different for the two IgG molecules studied, as would be expected for two monoclonal immunoglobulins. They bind strongly to their ligands and low pH conditions are necessary to elute them after immunosorption.

IgG λ Lac. and IgG λ Ler. behave like auto-antibodies. In the circulating blood they are inactive and at least partly linked to the circulating chylomicra from which they were extracted as well as to other lipoproteins and possibly albumin in case Lac. As such they are similar to the auto-antibodies found in AIH. Indeed in AIH, with the exception of some myeloma cases, the antibody activity is not detected in whole serum and has to be activated and purified as in the present instances.

TABLE 5. Auto-immune hyperlipidemia in myeloma

AIH type	Serum	Author	Antibody	Reactive antigen*	Antigen determinant
Anti-lipoprotein	Ger.	Beaumont *et al.* (1970a)	IgA κ	H (100%), AN, HDL, LDL, VLDL	Pg
	Sor.	Beaumont *et al.* (1970a)	IgA κ	H (100%), AN, HDL, LDL, VLDL	Pg
	God.	Lewis & Page (1965)*	IgA κ	H (100%), AN, HDL, LDL, VLDL	Pg
	Sav.	Beaumont *et al.* (1970a)	IgG κ	H (5%), HDL, LDL	AS
Anti-enzyme	Ed.	Glueck *et al.* (1972)	IgG κ	Heparin, heparin lipase	—†
	Sab.	Beaumont & Lemort (1970)	IgA κ	Heparin, heparin lipase	—†
Nephrotic syndrome	Lac.	Present study	IgG λ	H, AN, VLDL, HSA	
	Ler.		IgG λ	H, LDL	

* The reactive antigen was typed through the courtesy of Mrs Lena Lewis, who sent us a sample of serum. H = human, with frequency in parentheses. AN = animal. HDL = high density lipoproteins. LDL = low density lipoproteins. VLDL = very low density lipoproteins.

† The antigen determinant was different for cases Ed and Sab.

In previous studies on AIH in monoclonal gammopathies, several types of antibodies which react with different antigenic determinants were described (Table 5). By their type and their specific activities the IgG λ Lac. and IgG λ Ler. are still different. However, as was noticed for several of the AIH antibodies already known, they react with LDL, VLDL and chylomicra. They may therefore fit into the general concept in which AIH may be the result of the inhibition by an antibody of one of the numerous steps of the lipolytic process. In this view IgG λ Lac. and IgG λ Ler. would interact with one of the substrates of lipolysis and not directly with an enzyme. Moreover, the interaction of IgG λ Lac. with serum albumin, which was not noticed in other AIH cases, would also impair the lipolysis of triglycerides by blocking the main serum acceptor of free fatty acids. It is noteworthy that in case Lac. massive hypertriglyceridaemia was found at times, which was relieved by an albumin infusion, after which there was less extractable antibody activity in the serum. It is also

noteworthy that in case Ler., in which the antibody was anti-LDL and not anti-albumin, the hyperlipidaemia pattern was different, with a predominant hypercholesterolemia and only slight elevation of the triglyceride level.

It is foreseeable that several types of AIH will be found in the nephrotic syndrome in relation to the activity of antibodies differing from IgG λ Lac. and IgG λ Ler. The latter cases are good examples because the active Ig was monoclonal and was present in the serum in suitable concentration, thus giving a better opportunity to recover a sizeable amount of antibody. Among the types which may presumably be found in association with a nephrotic syndrome, it is likely that many may be of the anti-enzyme type, unlike cases Lac. and Ler. As a matter of fact, an inhibition of the plasma post-heparin lipase activity found in nephrosis (Yamada & Matsuda, 1970) which cannot easily be explained by anti-lipoprotein antibodies. However, this will have to be carefully studied because a low release of fatty acids in a test tube may be induced either by an insufficient amount of lipase, an inhibition of lipase, a blocking of the substrate, or by blocking the acceptor albumin.

If hyperlipidaemias associated with the nephrotic syndrome are AIH, the relationship between AIH and the renal lesions must be investigated. It is possible that the circulating antigen–antibody complexes produced in a variety of AIH may cause renal damage. However, with the exception of nephrotic syndromes renal damage is infrequent in hyperlipidaemia and not constant in AIH, although atherosclerosis, another tissue damage which may be induced by immune complexes, is very frequent in AIH (Beaumont & Beaumont, 1973) and was present in case Ler. On the other hand, if immune deposits were found in case Lac. they were not obvious in case Ler. The occurrence in the same individual of either renal lesions or atherosclerosis, or both, probably depends on the type of circulating antigen–antibody complexes. This has already been seen in experimental immune complex disease (Dixon *et al.*, 1961; Germuth, Senterfit & Pollack, 1967a).

It is felt that the antibody activity of IgG λ Lac. and IgG λ Ler. support the hypothesis that hyperlipidaemia in the nephrotic syndrome could be AIH. In this case the renal lesions could be secondary to the deposition either of the immune complexes involved in the AIH process or of other immune complexes, as it is well known that in auto-immune disease a variety of antibodies may occur in the same individual.

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