Combined IgG4 κ and IgG1 λ deposition in the glomerular and tubular basement membrane accompanied by autoimmune neutropenia (AIN) and immune thrombocytopenia (ITP)

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Abstract A 48-year-old male was admitted to our hospital with nephrotic syndrome. Light-microscopic examination of a renal biopsy specimen showed almost normal glomerular appearance, however, immunofluorescence examination revealed linear and granular IgG deposits on the glomerular basement membrane (GBM), accompanied by slight IgG deposition in the tubular basement membrane (TBM). Further investigation of the IgG subclass and light chain staining revealed that the glomerular deposits were composed of IgG1 and IgG4, with both κ and λ light chains, while the tubular deposits were composed of only IgG4 and κ light chains. The electron-microscopic findings of small granular deposits in the GBM and TBM closely resembled those of light and heavy chain deposition disease (LHCDD). Immunoelectron microscopy confirmed the presence of κ and λ chains in the GBM and TBM, however, only significant κ chain deposition was found in the TBM. There was no evidence of monoclonal gammopathy. Clinically, the patient subsequently developed neutropenia and thrombocytopenia associated with the presence of anti-neutrophil antibody and anti-GPIIb/IIIa antibody-producing B cells in the blood.

Oral steroid administration was initiated, which led to amelioration of the neutropenia, thrombocytopenia and proteinuria. This may be a very rare case of combined IgG4 κ and IgG1 λ deposition disease accompanied by autoimmune neutropenia (AIN) and immune thrombocytopenia (ITP) suggestive of biclonal immunoglobulin deposition disease (BIDD). Investigation of the IgG subclass and of the light chains was useful for recognizing the clonality of the immunoglobulin deposits in the kidney.

Keywords Nephrotic syndrome · Biclonal immunoglobulin deposition disease · IgG subclass · Light chain · Autoimmune neutropenia · Immune thrombocytopenia

Introduction

Monoclonal immunoglobulin deposition disease (MIDD) is associated with dysproteinemias, including monoclonal gammopathy of undetermined significance (MGUS) or multiple myeloma in some cases [1, 2]. The kidneys are the most commonly affected organs in MIDD [3]; renal histological findings reveal monoclonal light and/or heavy chain deposits in the glomerular, tubular and vascular membranes. Usually, MIDD is induced by the deposition of a single type of monoclonal immunoglobulin in organs, and no case with biclonal or oligo-clonal immunoglobulin deposition in the kidney has been reported so far.

Autoimmune neutropenia (AIN) is characterized by an absolute neutrophil count of less than 1500/ μ L in the presence of autoantibodies directed against neutrophil antigens, and can occur as a primary disease or secondary to various underlying diseases [4]. Recently, AIN was reported in a multiple myeloma patient accompanied by

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clonal proliferation of T cells, suggesting the possible existence of a relationship between monoclonal B cell and T cell lineages and AIN [5]. Immune thrombocytopenia (ITP) is an acquired hematologic condition caused by platelet-associated antibody [6]. It may occur in association with various diseases, including systemic lupus erythematosus (SLE) and human immunodeficiency virus (HIV) infection, or as a primary or idiopathic condition in the absence of any underlying disease. A few cases of ITP associated with multiple myeloma have also been reported [7].

Herein, we reported a very rare case of nephrotic syndrome accompanied by AIN and ITP, which was caused by unusual immunoglobulin deposits clinically resembling MIDD. Combined IgG4 κ and IgG1 λ deposition was detected in the GBM, although the TBM showed significant deposition of only IgG4 κ , suggesting the possibility of biclonal immunoglobulin deposition disease (BIDD).

Case report

Clinical history and laboratory data

A 48-year-old male was admitted to our hospital with nephrotic syndrome. He had an operation for Warthin's tumor of the right parotid gland at the age of 38. He had been suffering from chronic urticaria during the last 15 years, with allergic tendencies to several drugs. No urinary abnormalities were detected until one year before the onset. He visited a local hospital due to common cold-like symptoms, where proteinuria was detected. He was referred to our hospital for the management of nephrotic syndrome.

Laboratory examination (Table 1) revealed nephrotic-range proteinuria (14.2 g/gCr) with slight microhematuria. Examination of the blood chemistry revealed hypoproteinemia and hypoalbuminemia. His serum creatinine level was normal. Bence-Jones protein was not detected in the urine. Immunoelectrophoresis of the serum and urine disclosed no monoclonal immunoglobulin in either the serum or urine. Plasma immunofixation electrophoresis also did not show any monoclonal light or heavy chains. Serologic tests for antinuclear antibodies, cryoglobulin, and viral antigens (HIV, hepatitis B virus, and hepatitis C virus) were all negative. The plasma levels of immunoglobulins were relatively low presumably due to the massive proteinuria, with the exception of those of IgA. The serum complement levels were normal. There were no abnormal autoantibodies suggestive of autoimmune disease or vasculitis.

Kidney biopsy findings

Renal biopsy was performed to identify the underlying kidney disease causing the nephrotic syndrome. The

glomeruli were of almost normal appearance and showed no mesangial or endocapillary proliferations. No apparent sub-epithelial deposits or spike formations were detected in the glomerular capillary walls, however, careful observation revealed focal and segmental small irregularity of the glomerular capillary walls in limited regions on light microscopy (Fig. 1). Light microscopic examination revealed no significant thickening or depositions in the TBM.

Immunofluorescence microscopy revealed linear and granular IgG stainings in the glomerular capillary walls. Small granular C3 deposition was detected along the glomerular capillary walls, similar to the case for IgG, however, the staining intensity was weak (Fig. 2). IgG subclass staining revealed that the glomerular deposits were composed of IgG1 (2+) and IgG4 (3+). On the other hand, the TBM showed only weak staining for IgG4 (Fig. 3). Intensely positive staining for both κ and λ light chains was observed in the glomerular capillary walls corresponding to the deposition of IgG. In addition, careful observation revealed weak staining for only the κ light chain, but not for the λ chain, in the TBM (Fig. 3).

Electron microscopy revealed linear electron-dense deposits along the lamina densa and fine granular deposits in the GBM. There was no typical sub-epithelial electron-dense deposition suggestive of membranous glomerulonephritis (Fig. 4a). Similar fine granular deposits were also present on the epithelial aspect of the TBM (Fig. 4b). The electron-microscopic findings resembled those of light or heavy chain deposition disease (LHCDD), but apparently differed from those of membranous nephropathy (MN).

To elucidate the precise sites of deposition of the κ and λ light chains in the GBM and TBM, we then performed immunoelectron microscopy [8]. Briefly, ultrathin sections were prepared from the glutaraldehyde-osmium-fixed tissue block and pretreated with 10 % hydrogen peroxide for 5 min. The sections were incubated with primary antibodies for κ and λ light chains (DAKO) and then with the secondary antibody (goat anti-rabbit IgG antibody conjugated with 10 nm gold particles, Biocell International), followed by uranium staining. Sections without the primary antibody were used as control. Both κ and λ chains were observed in the GBM, but only significant κ chain deposition was found in the TBM. Although weakly positive staining for λ chain was also found in the TBM, it could not be distinguished from the high background staining. No significant difference in the location of the κ and λ chains in relation to the linear or granular deposits was demonstrated on immunoelectron microscopy (Fig. 5).

Clinical course after the diagnosis of the kidney disease

A bone marrow biopsy was performed after the renal biopsy to obtain evidence of monoclonality or

Table 1 Laboratory findings on admission

Urinalysis		Hematology	
Specific gravity	1.023	Hemoglobin	14.6 g/dL
Occult blood	±	Hematocrit	42.9 %
Protein	4+	White blood cell	3450/μL
Sediments		Neutrophil	65.5 %
Red blood cells	5–9/HPF	Eosinophil	3.8 %
Proteinuria	14.2 g/gCr	Platelet	19.1 × 10 ⁴ /μL
Bence-Jones protein	(–)		
Serum biochemistry		Immunological chemistry	
TP	5.1 g/dL	Rheumatoid factor	<10 U/mL
Alb	2.4 g/dL	Antinuclear antibody	<×20
AST	24 U/L	IgG	710 mg/dL
ALT	17 U/L	IgA	355 mg/dL
LDH	232 U/L	IgM	75 mg/dL
γGTP	34 U/L	IgE	1370 U/mL
BUN	8.4 mg/dL	C3	133.6 mg/dL
Cr	0.67 mg/dL	C4	34.7 mg/dL
UA	8.0 mg/dL	CH50	52.2 U/mL
Na	140 mEq/L	Cryoglobulin	(–)
K	3.7 mEq/L	M protein	(–)
Cl	107 mEq/L	MPO-ANCA	<10 EU
TG	298 mg/dL	PR3-ANCA	<10 EU
TC	236 mg/dL	Anti-GBM	<10 EU
HDL-C	32 mg/dL	CRP	0.10 mg/dL
HbA1c (NGSP)	5.4 %		

HPF high-power field, TP total protein, Alb albumin, AST aspartate amino transferase, ALT alanine amino transferase, LDH lactate dehydrogenase, γGTP γ glutamyltransferase, BUN blood urea nitrogen, Cr creatinine, UA uric acid, TG triglyceride, TC total cholesterol, HDL-C high density lipoprotein cholesterol, HbA1c hemoglobin A1c, MPO-ANCA myeloperoxidase anti-neutrophil cytoplasmic antibody, PR3-ANCA proteinase 3 anti-neutrophil cytoplasmic antibody, Anti-GBM anti-glomerular basement membrane antibody, CRP C-reactive protein

Fig. 1 Light microscopic findings of the kidney biopsy. **a** The glomeruli were of almost normal appearance, without any mesangial or endocapillary proliferations (PAS staining). **b** No apparent sub-epithelial deposits or spike formations were detected in the glomerular capillary walls, but careful observation revealed a focal and segmental small irregularity of the glomerular capillary walls in limited regions (arrows) (PAM staining)

Fig. 2 Routine immunofluorescent findings of the kidney biopsy. Linear and granular staining of IgG in the glomerular capillary walls, with a very slight linear deposition of IgG was noted in the TBM. Weak staining for IgA and IgM was detected mainly in the mesangium. Fine granular C3 deposition was detected along the glomerular capillary walls, similar to the case for IgG, but the staining intensity was weak

proliferation of immunoglobulin-producing cells, which revealed no abnormal proliferation of plasma cells (plasma cells: 2.4 %). During the course of hospitalization, the

patient eventually developed severe urticaria and was treated with 0.5 mg of betamethasone and 4 mg of D-chlorpheniramine maleate. Moderate neutropenia, with an

Fig. 3 IgG subclass and light chain κ and λ staining. The glomerular deposit was composed of IgG1 (2+) and IgG4 (3+). Subtle staining for IgG2 and IgG3 was seen. On the other hand, in the TBM, only a weak staining for IgG4 was noted. Intense staining for both κ and λ light chains was observed in the glomerular capillary walls. Only the κ light chain was detected in the TBM

Fig. 4 Electron-microscopic findings of the GBM and TBM. **a** The GBM revealed linear electron-dense deposits along the lamina densa (white arrow) and fine granular deposits, mainly on the sub-epithelial side of the GBM, showing a circular or ring-shaped appearance (white arrow head). There were no typical sub-epithelial electron-dense deposits. Podocyte foot process effacement was seen. **b** Similar fine granular deposits were also present on the epithelial side of the TBM (black arrows). The granular deposits on both the GBM and TBM predominated on the epithelial side of the basement membrane

absolute neutrophil count of greater than 500/ μ L, but lower than 1000/ μ L, occurred during the hospitalization; therefore, the anti-allergic agent cetirizine that the patient had been taking regularly was immediately stopped. Furthermore, because of sustained neutropenia and steroid-induced psychosis, betamethasone and D-chlorpheniramine maleate were also stopped after 6 months. No significant treatments were administered thereafter. The urinary protein excretion decreased, but persisted at the level of 5–6 g/gCr, accompanied by mild edema of the lower extremities. The white blood cell count decreased to around 500/ μ L. A repeat bone marrow aspiration was performed in November 2009, however, the cause of the neutropenia could not be determined. The neutropenia persisted despite the administration of granulocyte colony-stimulating factor (G-CSF), sometimes associated with fever. Platelet transfusions were administered when the patient's platelet count dipped below 20000/ μ L. Both anti-neutrophil antibody and anti-GPIIb/IIIa antibody-producing B cells [9] were detected in the peripheral blood in April 2011 (two years after the diagnosis of nephrotic syndrome), and the patient was diagnosed as having AIN and ITP. Immunosuppressive treatment with prednisolone 40 mg/day was started in July 2012 to control the AIN and ITP (Fig. 6). After the start of this treatment, the urinary protein excretion decreased from 1 to 0.2 g/gCr. There was no increase in the serum creatinine level during the follow-up period.

Discussion

Herein, we report a patient with atypical immunoglobulin deposition disease resembling LHCDD. On electron microscopy, non-organized fine granular deposits were detected in the GBM and TBM, similar to the findings in LHCDD, but apparently differed from those in MN. Furthermore, two types of deposits were observed on the GBM; linear and fine granular. Hence, the glomerular deposition probably represented combined monoclonal IgG (IgG4 κ and IgG1 λ) deposition, suggesting BIDD. However, no significant difference in the localization of the κ or λ chain was demonstrated in the linear or granular deposits in the GBM or TBM on immunoelectron microscopy.

Plasma cell dyscrasias are often detected after the diagnosis of MIDD [10]. At the time of the renal biopsy, approximately 30 % of renal MIDD patients have no detectable monoclonal protein in the serum or urine [2]. Renal histological findings reveal monoclonal light and/or

Fig. 5 Immunoelectron microscopic findings of GBM and TBM for κ and λ light chains

Fig. 6 The clinical course of the patient. *PSL* prednisolone, *G-CSF* granulocyte colony-stimulating factor, *Cr* serum creatinine concentration, *Neut* neutrophil count, *Plt* platelet count, *TP* total protein, *Alb* albumin, *Upro* urine protein/creatinine ratio

heavy chain deposits in the glomerular, tubular and vascular membranes. MIDD is classified into three types: light chain deposition disease (LCDD), heavy chain deposition disease (HCDD) and LHCDD. Lin et al. [11] described the composition of the deposits in 34 cases of MIDD, including 23 cases of LCDD, 5 of LHCDD, and 6 of HCDD. Our patient showed no clinical evidence of monoclonal gammopathy in the serum or urine, or of monoclonality/proliferation of immunoglobulin-producing cells in the bone marrow. However, the kidney biopsy findings strongly suggested LHCDD.

In our case, two different types of IgG (IgG4 κ and IgG1 λ) were deposited in an unusual manner, and we speculated that this was a case of biclonal IgG4 κ and IgG1 λ deposition disease. Biclonal gammopathy has been reported in various hematological diseases, such as monoclonal gammopathies and lymphoproliferative diseases including lymphoma and macroglobulinemia [12, 13]. Biclonal gammopathy accounts for around 1 % of monoclonal gammopathies, and many patients with biclonal gammopathy have IgG and IgA components (53 %), while less than 10 % of patients have two IgG components [13]. Furthermore, a case of symptomatic myeloma developed after the diagnosis of biclonal gammopathy was reported [13]. To our knowledge, no case of biclonal immunoglobulin deposition in the kidney has ever been reported. It is quite possible that this case represents a very rare case of BIDD in the kidney.

It is of interest that AIN and ITP occurred after the onset of the nephrotic syndrome in our patient. There are no previous case reports of MIDD complicated by AIN and ITP. Recently, Aryal et al. [5] reported the first case of a patient with multiple myeloma who developed AIN, in which the T cell receptor γ chain gene rearrangement indicated the presence of a clonal T cell proliferation, suggesting the possible existence of a relationship between monoclonal B cell and T cell lineages, and AIN. In addition, there have been only rare reports of ITP developing in patients with multiple myeloma [7]. The monoclonal protein itself might lead to AIN and ITP, although it could not be undetected in the serum or urine in our patient. Interestingly, the urinary protein excretion was reduced before the administration of immunosuppressive treatment. Furthermore, the level of proteinuria was not necessarily relevant to the severity of the neutropenia or thrombocytopenia. The relationship between the BIDD in the kidney and the serum autoantibodies against neutrophils and platelets seen in the present case may serve as an interesting subject for further investigation in the future. Longer follow-up is required to determine the implications of the relationship between renal and hematological disorders and the possible development of hematological malignancy in such patients.

Conclusions

We herein report an unusual case of IgG subclass and light chain deposition (IgG4 κ and IgG1 λ) in the kidney, possibly suggestive of BIDD, who subsequently developed of AIN and ITP, after the onset of the renal disease. This is a very rare case of atypical MIDD, and staining for the IgG subclass and κ and λ light chains was useful for recognizing the clonality of the immunoglobulin deposits in the kidney. Further case reports and clinical investigations are required to clarify the prevalence and significance of BIDD.

Conflict of interest The authors have declared that no conflict of interest exists.

References

1. Korbet SM, Schwartz MM. Multiple myeloma. *J Am Soc Nephrol.* 2006;17:2533–45.
2. Buxbaum J, Gallo G. Nonamyloidotic monoclonal immunoglobulin deposition disease. Light-chain, heavy-chain, and light- and heavy-chain deposition diseases. *Hematol Oncol Clin N Am.* 1999;13:1235–48.
3. Stratta P, Gravellone L, Cena T, Rossi D, Gaidano G, Fenoglio R, Lazzarich E, Quaglia M, Airoldi A, Bozzola C, Monga G, Valente G, Canavese C, Magnani C. Renal outcome and monoclonal immunoglobulin deposition disease in 289 old patients with blood cell dyscrasias: a single center experience. *Crit Rev Oncol Hematol.* 2011;79:31–42.
4. Akhtari M, Curtis B, Waller EK. Autoimmune neutropenia in adults. *Autoimmun Rev.* 2009;9:62–6.
5. Aryal MR, Bhatt VR, Tandra P, Krishnamurthy J, Yuan J, Greiner TC, Akhtari M. Autoimmune neutropenia in multiple myeloma and the role of clonal T-cell expansion: evidence of cross-talk between B-cell and T-cell lineages? *Clin Lymphoma Myeloma Leuk.* 2014;14:e19–23.
6. Cines DB, Blanchette VS. Immune thrombocytopenic purpura. *N Engl J Med.* 2002;346:995–1008.
7. Gupta V, Hegde UM, Parameswaran R, Newland AC. Multiple myeloma and immune thrombocytopenia. *Clin Lab Haematol.* 2000;22:239–42.
8. Herrera GA, Sanders PW, Reddy BV, Hasbargen JA, Hammond WS, Brooke JD. Ultrastructural immunolabeling: a unique diagnostic tool in monoclonal light chain-related renal diseases. *Ultrastruct Pathol.* 1994;18:401–16.
9. Kuwana M, Okazaki Y, Kaburaki J, Ikeda Y. Detection of circulating B cells secreting platelet-specific autoantibody is useful in the diagnosis of autoimmune thrombocytopenia. *Am J Med.* 2003;114:322–5.
10. Ronco P, Plaisier E, Mougenot B, Aucouturier P. Immunoglobulin light (heavy)-chain deposition disease: from molecular medicine to pathophysiology-driven therapy. *Clin J Am Soc Nephrol.* 2006;1:1342–50.
11. Lin J, Markowitz GS, Valeri AM, Kambham N, Sherman WH, Appel GB, D'Agati VD. Renal monoclonal immunoglobulin deposition disease: the disease spectrum. *J Am Soc Nephrol.* 2001;12:1482–92.
12. Toth Z, Sipos J. Biclonal primary plasma cell leukemia. *Pathol Oncol Res.* 1998;4:48–51.
13. Kyle RA, Robinson RA, Katzmann JA. The clinical aspects of biclonal gammopathies. Review of 57 cases. *Am J Med.* 1981;71:999–1008.