Progressive glomerulopathy with unusual deposits of striated structures: a new disease entity?

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Abstract

A 68-year-old man developed proteinuria and renal insufficiency. A renal biopsy showed mesangial proliferation and double contour in almost all glomeruli. Congo red staining for amyloid was negative. Immunofluorescence microscopy revealed no deposition of immunoglobulins. Electron microscopy showed unusual deposits of striated structures mainly in the subendothelial space and the mesangium. These deposits contained regularly stacked straight electron-dense bands. Microfilament-like deposits were also observed. The patient did not respond to steroid therapy and developed end-stage renal disease. All known disease entities with non-amyloid non-immunoglobulin-derived organized glomerular deposits were excluded. Progressive glomerulopathy in our patient might be a new disease entity.

Keywords: glomerulopathy; organized deposits; non-amyloid non-immunoglobulin-derived

Introduction

Until now, various organized glomerular deposits have been observed by electron microscopy in patients with renal diseases. These deposits are classified into three groups: Congo red-positive amyloid, non-amyloid immunoglobulin-derived deposits and non-amyloid non-immunoglobulin-derived deposits [1–3] (Table 1). The first group includes various types of amyloidosis. The second group includes cryoglobulinaemias, monoclonal gammopathies, systemic lupus erythematosus and immunotactoid glomerulopathy/fibrillary glomerulonephritis. The third group includes nail–patella syndrome, collagencefibrotic glomerulopathy, inflammatory glomerular diseases, diabetic nephropathy, fibronectin glomerulopathy and other glomerular diseases.

In this report, we describe unusual non-amyloid non-immunoglobulin-derived organized glomerular deposits in a patient with progressive renal disease. The deposits contained regularly stacked straight electron-dense bands. To our knowledge, such deposits have not been described in the literature.

Case report

A 68-year-old man was admitted to our hospital in March 2006 because of proteinuria and oedema. His family history and past history were unremarkable except for treated hypertension for 4 years. His proteinuria was first detected in another clinic in 2003, but detailed examinations had not been performed.

On admission, his blood pressure was 150/80 mmHg. A physical examination showed no abnormalities in the lungs, heart and abdomen. There was oedema of the lower limbs. Total 24-h urinary protein level was 1.4 g, and urine sediments showed 10–19 red blood cells/high power field. The haemoglobin was 12.9 g/dL, the white blood cell count 8700/μL and the platelet count 226 000/μL. The serum total protein was 6.5 g/dL, albumin 3.9 g/dL, blood urea nitrogen 30.0 mg/dL, creatinine 1.3 mg/dL, aspartate aminotransferase 21 U/L, alanine aminotransferase 21 U/L, total cholesterol 188 mg/dL, fasting blood sugar 95 mg/dL and haemoglobin A1c 5.2%. Serum IgG was 992 mg/dL, IgA 249 mg/dL, IgM 50 mg/dL, CH50 48.2 U/mL, C3 102.6 mg/dL and C4 36.6 mg/dL. Antibodies to hepatitis C virus, antinuclear antibodies and circulating immune complexes were negative. Serum cryoglobulin and monoclonal protein and urinary Bence–Jones protein were not detected. Serum type III procollagen peptide was 0.5 U/mL (normal 0.3–0.8 U/mL). There were no abnormalities on a chest roentgenogram and an electrocardiogram.

Light microscopy of renal biopsy specimens revealed moderate mesangial proliferation and double contour in almost all glomeruli (Figure 1). On trichrome-, periodic acid Schiff- and silver methenamine-stained sections, materials corresponding to the organized deposits described below...
were not stained. In the interstitium, mild lymphocyte infiltrations and mild tubular atrophy were found. There was mild hyalinosis of small arteries. Congo red staining for amyloid was negative. For routine immunofluorescence studies, frozen sections were stained with fluorescein isothiocyanate (FITC)-conjugated rabbit antisera to human IgG (\(\gamma\)-heavy chain), IgA (\(\alpha\)-heavy chain), IgM (\(\mu\)-heavy chain), \(\kappa\), \(\lambda\), C3, C1q and fibrinogen (DakoCytomation, Glostrup, Denmark). The results showed no significant staining for IgG, IgA, IgM, \(\kappa\), \(\lambda\) or fibrinogen. Positive staining for C3 and weakly positive staining for C1q were observed in the mesangium and along the glomerular capillary walls. Electron microscopy of lead citrate- and uranyl acetate-stained sections revealed unusual organized deposits of striated structures mainly in the subendothelial space (Figure 2) and the mesangium and partly in the subepithelial space (Figure 3) but not in the tubular basement membranes, interstitium or blood vessels. These deposits contained regularly stacked straight electron-dense bands. At a high magnification, the electron-dense bands were 10–12 nm in width with a center-to-center distance of 30–32 nm (Figure 2, inset). Microfilament-like deposits in a parallel arrangement containing partially striated structures were also observed in the subendothelial space (Figure 4). Microfilament-like deposits were occasionally seen mixed with deposits of striated structures (Figure 5). These organized deposits of striated structures and microfilament-like deposits were not clearly seen on phosphotungstic acid-stained sections, while collagen fibres with their typical periodicity were easily recognized on phosphotungstic acid-stained renal sections from a previously reported patient with collagenofibrotic glomerulopathy [4].

He received middle-dose steroid treatment (20 mg/day of prednisolone for 9 weeks), which was tapered gradually, and was followed up in an outpatient clinic from June 2006. He did not respond to steroid therapy and developed end-stage renal disease requiring haemodialysis in February 2009.
Immunohistochemical and immunofluorescence studies

Formalin-fixed and paraffin-embedded sections of renal biopsy specimens were processed for immunohistochemistry with mouse monoclonal antibodies to human collagen types I (Abcam, Tokyo, Japan), II (Daiichi Fine Chemical, Takaoka, Japan), III (BioLogo, Kronshagen, Germany), IV (Thermo, Cheshire, UK) and V (Daiichi Fine Chemical); rabbit polyclonal antibodies to human collagen types VI (Abcam) and XI (Santa Cruz Biotechnology, Santa Cruz, CA, USA); mouse monoclonal antibodies to human α-sarcomeric actin (Sigma, St. Louis, MO, USA), myosin (skeletal, slow) (Sigma) and lamin A/C (Abcam); and rabbit polyclonal antibodies to human myosin IIA (nonmuscle) (Sigma), myosin IIB (nonmuscle) (Sigma), lamin B1 (Abcam) and fibronectin (Cosmo Bio, Tokyo, Japan). The sections were stained using anti-mouse and anti-rabbit immunoglobins conjugated to a peroxidase-labelled polymer (DAKO, Carpinteria, CA, USA) and a liquid DAB substrate–chromogen system (DAKO). The specificity of antibodies were confirmed using skin biopsy specimens from a patient with systemic lupus erythematosus, muscle biopsy specimens from a patient with dermatomyositis, renal biopsy specimens from patients with diabetic glomerulosclerosis, renal autopsy specimens from a patient with collagenofibrotic glomerulopathy [4] and resected lung specimens containing hyaline cartilage from a patient with lung cancer.

Renal biopsy specimens were further examined with antibodies specific for γ-heavy chain domains. Cryostat sections were stained with mouse monoclonal antibodies to human IgG Fab containing C\(_{\text{H}1}\) domain (American Research Product, Belmont, MA, USA), IgG (Fc)-C\(_{\text{H}2}\) domain (AbD Serotec, Oxford, UK) and IgG (Fc)-C\(_{\text{H}3}\) domain (AbD Serotec), followed by FITC-conjugated anti-mouse IgG or anti-mouse IgM (Cappel, Aurora, OH, USA). Renal biopsy specimens from a patient with proliferative glomerulonephritis with monoclonal IgG deposits [5] were used as a positive control.

These immunohistochemical and immunofluorescence studies showed that glomerular deposits in biopsy specimens from the patient were not stained by antibodies to collagen types I–VI and XI, actin, myosin, lamin, fibronectin or IgG heavy chains.

Discussion

We reported a case of progressive renal disease with unusual organized glomerular deposits. These deposits were considered to be non-amyloid non-immunoglobulin-derived deposits, on the basis of the results of immunohistochemical stainings and immunofluorescence studies using antibodies specific for whole immunoglobulin heavy and light chains and for γ-heavy chain domains. Electron microscopy revealed deposits of striated structures containing regularly stacked straight electron-dense bands (10–12 nm in width) mainly in the subendothelial space and the mesangium and partly in the subepithelial space. Microfilament-like deposits were also observed mixed with deposits of striated structures in the subendothelial space. This suggests that microfilament-like deposits were partially organized precursory deposits of striated structures.

Non-amyloid non-immunoglobulin-derived glomerular deposits have been reported in various disorders [1–3] (Table 1). Nail–patella syndrome is a pleiotropic autosomal-dominant disorder characterized by dermatologic and musculoskeletal abnormalities. Patients with this syndrome show characteristic thickening of the glomerular basement membrane, including deposits of bundles of striated type III collagen fibres in the lamina densa [6]. These clinicopathological characteristics were inconsistent with those in our patient. Collagenofibrotic glomerulopathy is an idiopathic glomerular disease characterized by massive accumulation of atypical type III collagen fibrils in the mesangial matrix and subendothelial space and marked increase in serum type III procollagen peptide levels [7]. This glomerulopathy was excluded in our patient on the basis of negative staining for type III collagen and a normal serum level of type III procollagen peptide.
deposits of fibrin in inflammatory glomerular diseases, and the polymerized fibrin usually forms amorphous electron-dense masses. Rarely, there are fibrin tactoids with characteristic periodicity [3]. Fibrin tactoids were excluded in our patient because of negative staining for fibrinogen. Sohar et al. [8] reported cases of diabetic nephropathy with fibrils about 10 nm wide in some organs including the kidney. They suggested the name ‘diabetic fibrillosis’. Our patient had no clinical manifestations of diabetes mellitus. Moreover, typical histological findings of diabetic nephropathy were not observed. Fibronecetin nephropathy is a disease with an autosomal-dominant mode of inheritance characterized by glomerular fibronecetin deposits [3,9]. Patients with this nephropathy show fibrils with a diameter of 12–16 nm. This disease was excluded in our patient because of no family history and negative staining for fibronectin. Focal segmental glomerular sclerosis, malignant hypertension and haemolytic uraemic syndrome have been described with extracellular fibrils measuring 17–35 nm in diameter that are not associated with immunoglobulin deposition [1]. There were no clinicopathological findings of these disorders in our patient.

To our knowledge, only fibril-forming collagens (types I–III, V and XI) are known proteins that show a distance (D)-periodic banding pattern on the surface of molecules on electron microscopy in normal human tissues (where D=67 nm) [10]. This characteristic axial periodicity is due to the axial quarter-stagger packing arrangement of triple-helical collagen molecules. Immunohistochemical studies using specific antibodies suggest that fibril-forming collagens are unlikely sources of organized glomerular deposits in our patient. In addition, the deposits were not clearly seen on phosphotungstic acid-stained sections. Immunohistochemical stainings for basement membrane-forming collagen types IV and VI [10] were also negative.

From a morphological point of view, there is a general resemblance between the organized glomerular deposits of striated structures in our patient and striated crystalline bodies derived from animal contractile proteins and intermediate filament laminas [11–14]: observed crystalline bodies have highly regular axial banding repeats with the periodicity of ~14–45 nm on electron microscopy. However, these reported proteins are unlikely constituents of the glomerular deposits in our patient because in vivo formation of the reported crystalline bodies can occur under unphysiological conditions (for example, high concentrations of KCl and Mg2+, low pH and high pH). In addition, immunohistochemical stainings for actin, myosin and lamin were negative. Although the exact origin of the glomerular deposits of striated structures in our patient is uncertain at present, we suppose that the organized structures could be crystalline protein deposits.

As described above, all known disease entities with non-amyloid non-immunoglobulin-derived organized glomerular deposits were excluded in our patient. We suggest that progressive glomerulopathy in our patient might be a new disease entity with unusual deposits of striated structures containing presently unidentified substances. Accumulation of similar cases is needed to clarify clinicopathological features and the pathogenesis of this unique glomerulopathy.

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References

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