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Rapid progressive anti-GBM glomerulonephritis with multiple auto-antibodies

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ABSTRACT

Rapidly progressive glomerulonephritis (RPGN) is a clinical syndrome manifested by progressive loss of renal function in a short period. At renal biopsy, it is characterized by crescent formation. RPGN may be associated with the presence of circulating antibodies. We report a case of type IV RPGN [ANCA and Anti-glomerular basement membrane (anti-GBM) antibody disease], a severe disease causing a difficult to treat picture. Our case was complicated by severe thrombocytopenia due to the use of heparin and later on by thrombotic microangiopathy. These events occurred rapidly, making the clinical framing and management decisions very hard.

Implication for health policy/practice/research/medical education:

We report a case with the contemporaneous presence of multiple auto-antibodies (anti-GBM, ANCA, ANA, anti-PF4) and a clinical feature of rapidly progressive glomerulonephritis.

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Introduction

Rapidly progressive glomerulonephritis (RPGN) is a clinical syndrome manifested by features of glomerular disease in the urinalysis and by progressive loss of renal function in a short period (weeks or a few months). In renal biopsy, it is morphologically characterized by extensive crescent formation (1).

RPGN may be associated with the presence of circulating antibodies directed against the NC1 domain of the alpha 3 chain of type 4 collagen, expressed on the glomerular basement membrane (ab-GBM) (2) as in Good pasture disease or with anti-neutrophil cytoplasmic autoantibody (ANCA) as in microscopic vasculitis. Sometimes ANCA and anti-glomerular basement membrane (anti-GBM) antibody may be contemporaneously present, so-called

type 4 RPGN (3) causing a severe clinical picture; hemoptysis, resistant hypertension, alterations of bleeding tests (4,5) and rapid impairment of renal function.

We report a case of type IV RPGN [ANCA and anti-glomerular basement membrane (anti-GBM) antibody] (3) complicated by severe thrombocytopenia due to the use of heparin and later on by thrombotic microangiopathy (TMA). These events occurred in rapid succession, making the clinical framing and management decisions very hard.

Case Presentation

A 71-year-old Caucasian woman, in the early days of January 2019 had macroscopic hematuria and malaise treated with levofloxacin and ibuprofen. However, her

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clinical condition did not improve, and she subsequently developed lower extremity numbness, bilateral flank pain, fatigue and vomiting followed by anuria. On January 22, she was admitted to our hospital for acute renal failure. Physical examination revealed; lower-leg edema, body temperature 36.6°C, and arterial blood pressure 150/80 mm Hg. A chest X-ray showed bilateral pleural effusion. The renal function displayed a rapid impairment up to serum creatinine of 27 mg/dL and oligo-anuria (100 cc/24 h). Noticeably, she had no past medical history of renal disease, around 3 months before (October 2018), serum creatinine was 0.8 mg/dL. Urinalysis showed severe proteinuria (3.3 g/24 h), microscopic hematuria (3+) with dysmorphic erythrocytes. Anti-nuclear antibodies (ANAs) were positive 1:640 dilutions with a granular pattern, ANCA antibodies, anti-MPO, were positive (24 U.A/mL, normal value <10). Anti-GBM antibodies were detected in the blood sample (647 U/mL, normal values <7). Red blood cells 3.400.000 mmc, hemoglobin 10 g/dL, white blood cells 10.800 mmc, platelets 238.000 mmc. LDH was increased (324 U/L). The hemodialysis was started with enoxaparin for anticoagulation. The renal ultrasound showed normal-sized kidneys and a biopsy was carried out.

Light microscopy showed 18 glomeruli; all of them had large, circumferential, cellular crescents associated with inflammatory infiltrating cells and focal gaps of Bowman's capsule. The small vessel walls showed focal severe fibrinoid necrosis. A severe, diffuse inflammatory cellular infiltrate was also evident in the interstitium (Figure 1). Direct immunofluorescence showed linear staining of IgG (3+) along the glomerular basement membrane. A granular, mesangial C3 deposition was present as well. Linear staining for fibrinogen (++) was also present along the crescents (Figure 2). Ultrastructural investigation showed focal interruption of the basal glomerular membrane, fibrin tactoids and electron dense deposits, likely C3 deposition (Figure 3).

A high-resolution lung-CT did not display any signs of involvement. The patient received one g/d intravenous methylprednisolone for three days, followed by oral prednisone (1 mg/kg/d) and cyclophosphamide (1 mg/kg/d). Moreover, an intensive daily course of plasma exchange was started.

Before the first plasma exchange the patient developed severe thrombocytopenia (Figure 4), cyclophosphamide was drawn, and heparin-induced thrombocytopenia (HIT) was suspected. Detection of antibodies to PF4 in complex with heparin (anti-PF4) was requested although the pre-test probability score, assessed by the four T-test, was low (6). Anticoagulation with heparin was stopped, and anticoagulation during hemodialysis and plasma exchange was obtained with the inhibitor of factor Xa.

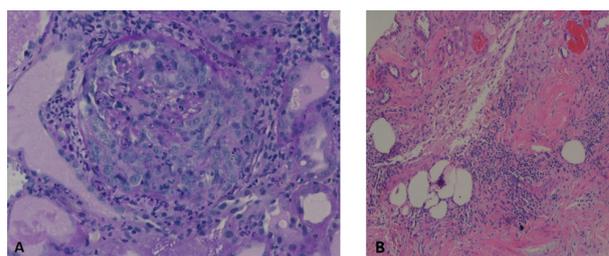


Figure 1. A large circumferential proliferative cellular crescent and an extensive dissolution of glomerular basement membranes coexists. The Bowman's capsule is fractured in the right side with continuity of changes between interstitium and capsule associated with a severe cellular proliferation (LM PAS x40). B The picture displaying a small arteria with extensive fibrinoid necrosis of the wall (H&E x40).

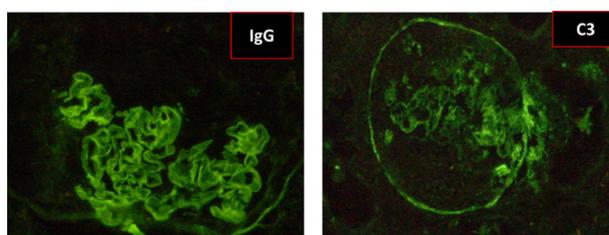


Figure 2. Linear staining along GBM for IgG, mesangial granular deposits of C3.

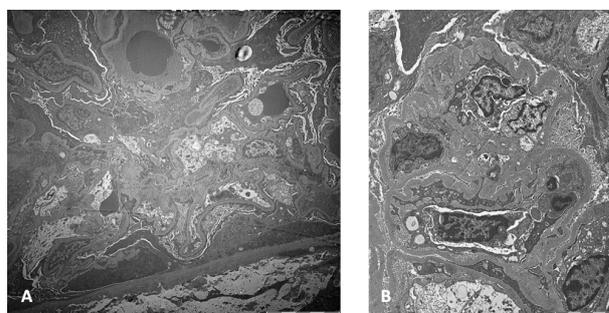


Figure 3. A electron microscopy show collapse of capillary walls of the remnant glomerular and fibrin tufts with interruption of GBM B Electron microscopy shows the presence of the electron dense deposits along the capillary walls.

In the following days, platelets fell up to 13.000 (U/ μ L), LDH was 330 IU/l, haptoglobin 8 mg/dL, serum bilirubin 2.78 mg/dL (direct 0.48 mg/dL), Hb of 9.5 g/dL and 3% of schistocytes were present at peripheral blood smear as well. The direct and indirect Coombs test was negative. On the base of laboratory data gathered, a diagnosis of TMA was made. In the hypothesis that an atypical hemolytic uremic syndrome could be present the activity of disintegrin-like and metalloproteinase with thrombospondin type 1 motif, 13 (ADAMTS13) by

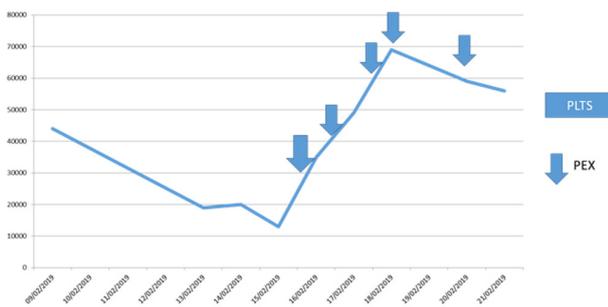


Figure 4. The time course of RPGN caused by anti-GBM glomerulonephritis in a patient with thrombocytopenia and ANCA. The platelet count began to decrease approximately two weeks after the start of HD. Plt began to arise after the second PEX and showed maximum increase at five treatment.

chromogenic enzyme-linked immunoassay (ADAMTS13-act-ELISA) was requested.

After five consecutive plasma exchange treatments, for total exchange of 1.5 volume per treatment, platelet count increased to 69.000 (U/μL), and the hemolysis indices normalized. The results of the ADAMTS13 were available some days later and the ADAMTS13 activity of 64% (reference range 70%–120%), ADAMTS13 inhibitors were negative. In contrast, the result of anti-PF4 IgG was positive. The hemodialysis was continued with an inhibitor of factor Xa while plasma exchange was discontinued at six treatments.

The patient was discharged after a few days, and she remained in chronic dialysis treatment and did not develop pulmonary symptoms.

Discussion

This case of RPGN is peculiar because of the contemporaneous presence of multiple auto-antibodies (anti-GBM, ANCA, ANA and anti-PF4) involved in pathogenesis and clinical outcome. Moreover, these antibodies significantly affected clinical management and therapeutic decisions.

The incidence of ANCA in anti-GBM antibody-related disease is reported as approximately 30%–40% (4). In this case, as usual, both antibodies were present at the diagnosis (7). However, the appearance of the anti-GBM may occur both before ANCA (7) and later after kidney injury due to pauci-immune origin of crescentic glomerulonephritis (3,8), and the renal outcome is anyhow poor. However, McAdoo et al (9) described a group of patients in which double-positive was associated with a better trend of renal function but burdened by high mortality. Double-positive patients exhibit features of both diseases (9). At histology, double-positive RPGN usually exhibits advanced renal damage: sclerotic glomeruli and tubulointerstitial fibrosis (10) though in our case the acute

damage was prevalent with cellular epithelial hyperplasia and with inflammation changes (fibrinoid necrosis). To have a pathogenetic framing, we can speculate that the extracapillary proliferation was due to anti-GBM and the fibrinoid necrosis of arterioles to ANCA. The coexistence of these two patterns of lesions is reported in patients with ANCA and anti-GBM indeed acute inflammation of renal vessels, other than glomerular capillaries, is not typical for the only anti-GBM glomerulonephritis, and suggests an ANCA origin (11). In this patient, the adverse prognostic factors at the diagnosis (anuria, high serum creatinine and focal gaps of Bowman's capsule) made end-stage renal disease as the more probable outcome.

Our clinical report was further complicated by thrombocytopenia (53 000 U/μL platelets on 13th day from the start of treatment) and by severe anemia which became the life-threatening problem. The presence of many aspects of autoimmunity (ANA, anti-GBM, ANCA anti-MPO) could suggest an autoimmune origin of the haematological complications (12). However, the ANA positivity was not associated with other typical features of Lupus. The second hypothesis was HIT. HIT typically is divided into type I and II. HIT-I is a mild, transient drop in platelet count, and is not associated with thrombosis, and is not considered clinically significant. HIT type II (HIT II) is a clinically significant syndrome due to antibodies to PF4 complexed to heparin (13). These antibodies can cause thrombosis along with thrombocytopenia. Presence of auto-antibodies is described in 20%-30% of patients in HD who are exposed to heparin for anticoagulation, but only 1%-3% developed the syndrome (14). Although the timing of presentation of the disease was late (>10 days) for a HIT II (15), we searched for anti-PF4 antibodies. Our patient did not develop thrombosis (lower limbs Doppler sonography was negative). The third hypothesis was cyclophosphamide associated toxicity, although the total dose administered was low (1 mg/kg/d for 10 days). A bone marrow biopsy was performed and no changes due to cyclophosphamide toxicity were revealed, but we suspended the alkylating agent temporarily. The fourth hypothesis was a direct side-effect due to plasma exchange (16), and we suspended the treatment after the first exchange.

Notwithstanding our therapeutic decisions, platelet count kept on reducing (up to 13 000 U/μL). It is noticeable that severe thrombocytopenia is rare due to HIT-II, and a suspicion of other causes arose (17). The negativity for Coombs tests and the presence of schistocytes addressed towards hemolysis with platelet consumption of intravascular origin, and a diagnosis of TMA was made.

TMA usually occurs in patients with autoimmune diseases (18). Several studies have reported a combination

of TMA and either anti-GBM antibody-related diseases (19, 20) or MPO-ANCA-related diseases (21,22). TMA is a pathological condition characterized by thrombosis of small vessels, intravascular breakdown of red blood cells, elevations in the levels of serum LDH and consumption of platelets. However, TMA is also associated with other clinical conditions, such as malignant hypertension, malignancies, post-transplantation and various types of glomerular injuries (23). Not surprisingly, an association between TMA and anti-GBM glomerulonephritis has been demonstrated as well (19,20). The cases were characterised by a renal limited anti-GBM disease complicated by fever and microangiopathic hemolytic anemia, without an apparent cause. Because a diagnosis of TMA was made, treatment with steroids and plasma-exchange was started, with partial remission was obtained.

ADAMTS13 activity plays a crucial role in classified TMA (24). ADAMTS13 activity is less than 5% of normal due to the presence of an autoantibody or inhibitor against ADAMTS13 and can be successfully treated with eculizumab for atypical HUS. There may be an etiological link between an ADAMTS 13-dependent mechanism and the development of TMA in patients with anti-GBM glomerulonephritis (18). In the absence of ADAMTS13: AC the UL-VWFMs produced in vascular endothelial cells and released into circulation excessively causing platelets to aggregate under high shear stress. The result is a luminal narrowing, hyperviscosity, and high flow rate of blood. However, this was not the case; in the current patient, ADAMTS-13 did not likely play a significant role because no decreases in the ADAMTS 13 activity were demonstrated. The patient presented in this report had normal ADAMTS13, preventing us from using anti-CD20 or anti- C5 antibodies.

Moreover, the presence of vasculopathy associated with TMA, such as occluded arterioles with fibrin plugs or schistocytes, was not confirmed in the renal biopsy of the patient. This is not surprising, however, since clinical phenotypes and pathological findings may not necessarily be confirmed simultaneously (25).

Our diagnosis was TMA associated with HIT-II, and we restarted patient treatment with an additional five plasma exchange and fresh frozen plasma in reinfusion. In the following days a significant improvement of platelets count was observed, and hemolysis index was normalized.

Conclusion

We report a patient with the contemporaneous presence of multiple auto-antibodies (anti-GBM, ANCA, ANA, anti-PF4) and a clinical feature of RPGN making a complicated case of dysregulation of immune processes. Moreover, this is to the best of our knowledge, the first report of positive glomerulonephritis associated with anti-PF4

positivity. Correct evaluation in the differential diagnosis is tremendously crucial for the choice of treatment.

Authors' contribution

DM and SF wrote the paper, treated and followed the patient and revisited the case. TVR reviewed samples and reported pathology results. ART was the electron microscopist who read TEM. AS and FM were the physicians of the patient and completed data and helped in writing the draft. All authors read and signed the final paper.

Conflicts of interest

There is no conflict of interest in this study.

Ethical considerations

Ethical issues including plagiarism, double publication, and redundancy have been completely observed by the authors. The patient gave the consent to publish as a case report.

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