

Efficacy of Rituximab in a Patient With Partial Clinical Remission and Persistent Circulating PLA2R-Ab



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INTRODUCTION

Membranous nephropathy (MN) is one of the leading causes of nephrotic syndrome in adults. Primary MN (PMN) is associated with a significant risk of developing end-stage renal disease, but clinical evolution is overall unpredictable.¹ Until recently, proteinuria was the main biomarker to assess severity and choose therapeutic indications,² and it is the primary criterion used to define remissions. Indeed, according to Kidney Disease: Improving Global Outcomes (KDIGO) guidelines, partial remission and complete remission are defined by proteinuria between 0.5 and 3.5 g per day (and less than 50% of the baseline value) and less than 0.5 g per day, respectively.³ However, proteinuria is an imperfect marker. In some patients, residual proteinuria may long persist in the absence of immunologic activity, because of tubulo-interstitial damage or secondary glomerular changes caused by prolonged disease, and conversely, in other patients with moderate levels of proteinuria, renal function may fast deteriorate.⁴

In 2009, Beck *et al.*⁵ found that 70% of adult patients with PMN produced autoantibodies against an antigen expressed on podocyte cells, the M-type phospholipase A2 receptor (PLA2R). There is accumulating evidence that high titers of PLA2R antibodies (PLA2R-Abs) are correlated with a lower risk of spontaneous or immunosuppressor-induced remission, a higher risk of nephrotic syndrome, and of progression to end-stage renal disease.^{6–8} Rituximab is effective in achieving immunological remission, which usually precedes clinical remission of proteinuria by several months.^{9,S1–S3} Detection and measurement of PLA2R-Abs therefore

play an important role in monitoring disease activity and treatment efficacy. However, there may be discrepancies between immunological and clinical parameters, and the KDIGO guidelines do not specify whether patients with clinical partial remission but persisting immunological activity should be re-treated with immunosuppressive agents despite the harmful effects of prolonged proteinuria and cardiovascular risk.³

We report here the case of a patient with persistent circulating PLA2R-Abs in the setting of prolonged partial remission and stable renal function, in whom we undertook a third line of immunosuppressive therapy.

CASE PRESENTATION

A 31-year-old man, without any past medical history, developed a nephrotic syndrome in December 2003 (proteinuria 8.2 g/d, serum albumin 2 g/dl) without hematuria or renal failure (serum creatinine 0.9 mg/dl and estimated glomerular filtration rate [Modification of Diet in Renal Disease formula] 99 ml/min) (Table 1, Figure 1). A kidney biopsy revealed a stage-2 MN. Antiproteinuric treatment with ramipril and amiloride was started. A first line of immunosuppressive treatment with alternating monthly cycles of corticosteroids and chlorambucil was administered, starting in June 2004 for 6 months. This treatment was complicated by azoospermia and neutropenia and was not effective.

One year later, the patient still presented with nephrotic syndrome (creatinine 0.9 mg/dl, proteinuria 7 g/d, serum albumin 2.5 g/dl). According to KDIGO recommendations, he received a second line of immunosuppressive treatment with cyclosporine 5 mg/kg per day for 18 months.^{S4} The patient achieved partial

Table 1. Laboratory results

Characteristics	November 2013	September 2014	March 2015	June 2016	August 2016	March 2017	June 2017
Creatinine (mg/dl) ($\mu\text{mol/l}$)	1.3	1.1	1.5	1.4	1.6	1.4	1.6
MDRD GFR (ml/min per m^2)	60	63	48	54	46	54	46
Proteinuria (g/d)	2	2	0.5	1.1	1.6	0.5	0.1
Serum albumin (g/dl)	3.2	3.0	3.0	3.0	2.9	3.8	4.0
PLA2R-Ab (ELISA)	112	124	122	98	194	40	0

ELISA, enzyme-linked immunosorbent assay; GFR, glomerular filtration rate; MDRD, Modification of Diet in Renal Disease; PLA2R-Ab, phospholipase A2 receptor antibodies.

remission of nephrotic syndrome within 3 months after initiating cyclosporine (creatinine 1.45 mg/dl, proteinuria 2.8 g/d, serum albumin 3.5 g/dl). Cyclosporine was progressively reduced and then withdrawn in August 2007.

The patient remained in partial remission of nephrotic syndrome with stable kidney function for the following 10 years. PLA2R-Ab was assessed for the first time by enzyme-linked immunosorbent assay (EUROimmun, Lübeck, Germany) in November 2013, and was positive at 112 RU/ml (Table 1).

Circulating PLA2R-Abs persisted for the next 3 years, PLA2R-Ab level was 194 RU/ml in August 2016. Then, contrary to KDIGO guidelines, despite the absence of nephrotic syndrome or rise of serum creatinine (serum albumin was 2.9g/dl and proteinuria was 1.6 g/d), a third line of immunosuppressive treatment was given with the aim to achieve complete clinical

remission. The patient received 2 infusions of rituximab (375 mg/m^2 per infusion) at 1-week interval. Because the patient developed leukopenia and infertility after chlorambucil treatment, we started with a relatively low dose of rituximab. Six months later, a complete clinical remission was achieved (serum albumin, 3.8 g/dl; proteinuria, 0.5 g/d), associated with partial immunological remission (PLA2R-Ab level dropped to 40 RU/ml). CD19+ B cells rose above 10 per mm^3 . Because of CD19+ B-cell repletion and the risk of relapse associated with persisting PLA2R-Abs,⁹ we decided to perform a second course of rituximab (1 g) in March 2017 with the aim of stopping the residual antibody production. Three months after this course, the patient maintained complete remission of nephrotic syndrome (albumin level 4.0 g/dl, proteinuria 100 mg/d) and PLA2R-Abs were for the first time undetectable. The patient remains in complete clinical and immunological remission 2 years later without any side effect of treatment.

DISCUSSION

PMN is an autoimmune disease caused by autoantibodies directed against podocyte antigens. The commonest antibody is directed against PLA2R. Indications of, and response to, therapy is traditionally assessed by using standard laboratory parameters, such as proteinuria, serum albumin, and serum creatinine levels. Based on KDIGO guidelines,³ our patient achieved partial remission in 2005, did not relapse, and thus did

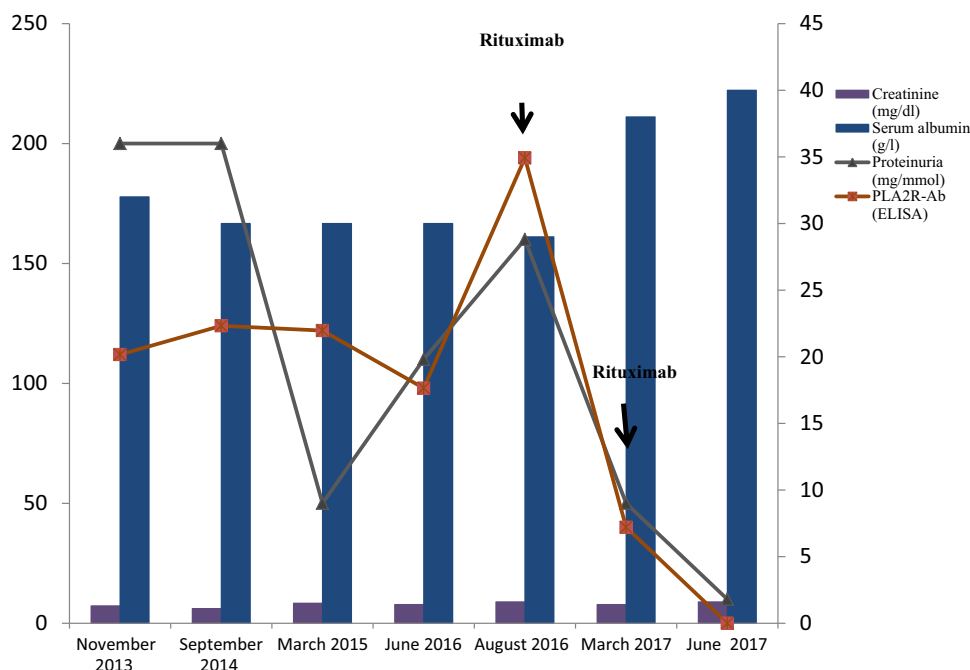


Figure 1. Evolution of creatinine, serum albumin, proteinuria, and anti-PLA2R-Ab. PLA2R-Abs are assessed by enzyme-linked immunosorbent assay (ELISA) and expressed as RU/ml.

not present any indication for use of an additional immunosuppressive therapy during the 12 years of follow-up. Here we show that this “wait and see” attitude should be reconsidered based on persistence of immunological activity. The pathogenic role of PLA2R-Abs is not clearly established, but antibody levels correlate with disease activity (Table 1). A high level of antibodies is a risk factor for development of nephrotic syndrome and end-stage renal disease, for failure to treatment, and recurrence after kidney transplantation.⁵⁵ Antibody levels commonly decrease before spontaneous or induced remissions and rise before relapses. Their persistence usually predicts poor clinical outcome.^{6–9} Clinical parameters (proteinuria, albumin level, and glomerular filtration rate) of our patient remained stable for more than 10 years, but these biomarkers are clearly imperfect and may underestimate smoldering kidney damage induced by this immunological disease (Table 2). Thus, because of persistent high levels of PLA2R-Ab, we feared subclinical, progressive renal damage with risk of evolution to end-stage renal disease.

For more than a decade, the anti-CD20 monoclonal antibody rituximab has been used in patients with MN, and data indicate that rituximab is an effective drug in promoting disease remission in patients with PMN and severe nephrotic syndrome.^{9,S2,S3,S6} An important, but frequently neglected advantage of rituximab is its major impact on the quality of life of patients. Patients receiving rituximab benefit from improvement of symptoms while they are not affected by toxicity of treatment, contrary to those treated with steroids, alkylating agents, or calcineurin inhibitors.^{S3–S7} Thus, we decided to treat this patient with rituximab therapy after more than 10 years of sustained partial remission.

De Vriese *et al.*^{S8} suggested a serologically based approach to manage patients with MN. They based the indication of treatment on classic parameters such as proteinuria and albumin level, but also on PLA2R-Ab titer. For the first time, the authors recommended to

treat patients with a high titer of PLA2R-Ab (>204 RU/ml) with immunosuppressive treatment, regardless of the level of proteinuria, and to evaluate therapy response on clinical remission and evolution of PLA2R-Ab titers. They also suggested that the goal of immunosuppressive treatment should be to decrease the level of antibodies by 90% after 6 months. Our case supports the PLA2R-Ab–driven therapy suggested by De Vries *et al.*,^{S8} which we applied to a patient with prolonged partial remission and immunological activity. We treated the patient with rituximab because of persistent high levels of PLA2R-Ab, even though antibody level remained less than 204 RU/ml.

We agree with the approach of De Vries *et al.*^{S8} and we think that treatment endpoint should be disappearance of immunological activity (Table 1). Ruggenti *et al.*^{S1} showed that the rate of clinical complete remission is higher when PLA2R disappear ($P < 0.0001$). Therefore, in our patient, we aimed at complete disappearance of antibodies and we hoped that he would subsequently achieve sustained complete remission. Optimal dosing of rituximab is still controversial, although already in 2010, a pharmacological study in nephrotic patients with PMN showed that B-cell depletion was shorter and that CD19+ B cells recurred before 90 days, whereas depletion continued over 180 days in patients with rheumatoid arthritis.^{S9} Because our patient developed leukopenia and infertility after chlorambucil treatment, we started with a relatively low dose of rituximab and we reinfused him 6 months later at a PLA2R-Ab level of 40 RU/ml. Although we could not retrieve archival sera before 2013, one can reasonably anticipate that the 2 lines of treatment with the protocol of Ponticelli and Glasscock¹ and cyclosporine decreased PLA2R-Ab titers, but the disease was still immunologically active during partial remission. De Vries *et al.*,^{S8} using the enzyme-linked immunosorbent assay test, suggested that the goal of immunosuppressive treatment should be to decrease the level of antibodies by 90% after 6 months. However, there is a gray zone between 2 RU/ml and 20 RU/ml in which a substantial number of samples remain positive by immunofluorescence. A 90% drop may fall into this gray zone. We agree that enzyme-linked immunosorbent assay should be used for treatment monitoring, but we prefer to use as treatment endpoint the disappearance of antibodies by immunofluorescence.

It is likely that the forthcoming KDIGO guidelines will integrate serology in the therapeutic algorithm of patients with MN and thus provide guidance for more personalized medicine as illustrated by this observation. Remaining questions regard PLA2R-Ab thresholds, the place of epitope spreading, and that of genetic

Table 2. Distinct teaching points

Phospholipase A2 receptor antibody (PLA2R-Ab) levels correlate with disease activity.
There may be discrepancies between immunological and clinical parameters.
Immunosuppressive treatment of patients in partial clinical remission should be considered because of renal and cardiovascular risk.
Rituximab can help achieve complete clinical and immunological remission in patients with partial remission.
Detection of PLA2R-Ab by immunofluorescence test (IIFT) is more sensitive than enzyme-linked immunosorbent assay when using the threshold of 20 RU/ml recommended by the manufacturer.
Reports suggest that the ultimate goal of the immunosuppressive treatment in PLA2R-Ab–positive patients should be complete disappearance of antibody by IIFT, but this should be confirmed by controlled prospective studies.

risk score. They will hopefully be solved by adaptive, serology-driven randomized controlled trials.

DISCLOSURE

All the authors declared no competing interests.

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SUPPLEMENTARY MATERIAL

Supplementary References.

Supplementary material is linked to the online version of the paper at www.kireports.org.

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