Factors affecting outcome and prognosis in membranous lupus nephropathy

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Abstract

Background. This study was designed to review the prognosis and the predictors of renal outcome in patients with membranous lupus nephropathy (MLN) with no or mild mesangial proliferation.

Methods. The medical records of patients (n = 66) with biopsy-proven MLN, WHO class VA, and class VB without any past history of proliferative lupus nephropathy (PLN) were reviewed retrospectively.

Results. The mean follow-up was 6.9 ± 0.2 years and renal survival at 5 and 10 years was 97 ± 2 and 88 ± 6%. Twenty-nine patients underwent a second renal biopsy during follow-up. Fourteen of these patients (21%) had lesions of PLN. Among them, four reached end-stage renal disease (ESRD) despite immunosuppressive treatment. The probability of a transition from MLN to PLN at 10 years was 35 ± 8%. Two other patients reached ESRD but did not have repeat renal biopsies and two had biopsy-proven progression to fibrosis. Independent risk factors associated with the doubling of creatinine were transition into PLN and the occurrence of a thrombosis during follow-up. The only predictor of ESRD was the haemoglobin level. PLN was not a predictor of ESRD but the efficient treatment of this form of lupus nephritis prevented that outcome.

Conclusions. With a long follow-up, our study noted a high frequency of transition into PLN in a large cohort of patients with MLN. Steroid usage was not predictive of outcome and did not affect renal survival, a result that must be qualified in light of the highly variable duration of treatments with steroids. The early screening and treatment of PLN is the main benefit of the close follow-up of patients with MLN. Progression to ESRD with only fibrosis remains a rare event.

Keywords: lupus nephritis; membranous lupus nephropathy

Introduction

The World Health Organization’s classification of lupus nephritis (LN) is in wide use by clinicians. Classes III and IV, respectively, defined by a segmental (<50%) or a diffuse endocapillary proliferation (>50%), have the worst natural history, but their prognosis has been considerably improved by immunosuppressive treatment. Membranous lupus nephropathy (MLN), class V in the WHO classification, was formerly subdivided into four groups ranging from the pure membranous nephropathy (Va) to the membranous nephropathy with diffuse superimposed proliferative lesions in the subclass Vd. As the subclasses Vc and Vd have a clinical course and a prognosis similar to the proliferative classes III and IV [1], the WHO classification was recently revised, and the subclasses Vc and Vd were reclassified in to classes III or IV depending on active necrotizing lesions. Class V now includes only subclasses VA and VB.

Pure or minimally proliferative MLN has a better outcome than classes III and IV [2,3]. Our multicentre study was designed to define the course and prognosis of LN classes VA and VB (pure MLN and MLN with superimposed mild mesangial proliferation) and to search for the predictors of outcome.
Patients and methods

Patients

This multicentre, retrospective clinicopathologic study evaluated 66 patients with MLN who underwent renal biopsies between January 1976 and January 1997 in seven Parisian hospitals. The renal biopsy of registers of each hospital was used to identify eligible patients. Medical records were reviewed in chronological order and the data extracted.

Selection criteria

All patients met the 1982 American Rheumatism Association (ARA) revised criteria for diagnosis of SLE—if not at the time of renal biopsy then during follow-up. Of those criteria of SLE, all patients had clinical evidence of renal disease defined by proteinuria equal or superior to 0.5 g/day or haematuria or leucocyturia, with or without renal failure. The start of the study for each patient was the day of the renal biopsy diagnosed MLN.

Pathologic criteria

The diagnosis of MN required the presence of diffuse basement membrane thickening on light microscopy and granular basement membrane deposits of IgG on fluorescence microscopy. Only patients with no (VA) or mild mesangial proliferation (VB) were included. Patients were excluded if MLN was associated with proliferative lesions (former classes Vc and Vd) and if the patients had been diagnosed previously with a proliferative form of LN (PLN, including subclasses III, IV, and former Vc and Vd). A pathologist unaware of the clinical courses of the patients reviewed the renal biopsies that had been processed for light microscopy.

Pathologic findings were classified using the four-stage developmental scale of the idiopathic MN: stage I, subepithelial deposits; stage II, spike formation; stage III, incorporation of the deposits into the glomerular basement membrane; and stage IV, disappearance of deposits. The number of glomeruli, sclerotic glomeruli, and glomeruli with hyaline deposits were counted. Arteriolosclerosis, tubular atrophy, interstitial inflammation, and fibrosis were scored 0–3 with 0 = absent, 1 = mild, 2 = moderate, 3 = marked. Final pathologic diagnosis was classified as MLN, MLN with intrarenal vascular lesions suggestive of anti-phospholipid syndrome, or MLN with predominant arteriosclerosis.

Clinical evaluation

Baseline clinical data included: age, sex, ethnicity, blood pressure, and weight. The cumulative number of ARA criteria, calculated at baseline, reflected disease severity.

The following laboratory values for the time of biopsy were analysed: serum creatinine, serum albumin, 24-h proteinuria, third component of serum complement C3 (classified either as in the normal range or decreased), the value of serum antinuclear antibody, the presence of serum antibodies to native undenatured DNA (dsDNA), the presence of serum antibodies to non-DNA nuclear and cytosolic antigens (Sm, nRNP, SSA, SSB), the presence of cryoglobulinemia, and of antiphospholipid antibodies (lupus anticoagulant and/or anticardiolipin antibody). Creatinine clearance was calculated using the Cockcroft and Gault formula.

Every year during follow-up, at least one assessment of serum creatinine, proteinuria, and albuminaemia was made. At the end of the follow-up, the same parameters were tested and recorded along with the date of the test. Clinical thromboembolic events were screened from baseline to the end of follow-up or to transition into a PLN. Results of serial kidney biopsies performed during follow-up were scored according to the WHO classification for LN. Treatment prescribed just after the diagnosis of MLN was subdivided into three classes: group 1, corticosteroids $\geq$ 1 mg/kg/day; group 2, corticosteroids <1 mg/kg/day (mainly 0.25–0.5 mg/kg/day); and group 3, no steroids and no other major immunosuppressive therapy.

Study outcomes

Major end points were either death or end-stage renal disease (ESRD) requiring chronic renal replacement therapy. Secondary end points included remission, doubling of serum creatinine, and transition to PLN. Prolonged remission was defined by a 24-h proteinuria $<$ 0.5 g/day, the absence of haematuria and leucocyturia and no relapse during follow-up.

Statistical analysis

We performed a univariate analysis using the $\chi^2$ test for qualitative variables (serum creatinine doubling, remission) and the Cox model for failure—point variables (renal death, transition into PLN). The following variables were examined: ethnicity, time elapsed between the diagnosis of SLE and MLN, at baseline, cumulative ARA score, renal characteristics (proteinuria, serum albumin, Cockcroft clearance, haematuria, hypertension), lupus biological activity (sedimentation rate, haemoglobin level, decreased or normal serum C3, logarithm of serum nuclear antibody, presence or absence of anti-ds-DNA), immunological status (presence or absence of Sm, SSA, SSB, RNP, aPL), proteinuria and serum albumin at 1 year of follow-up and thrombosis.

For multivariate modelling, qualitative and quantitative variables significantly associated with dependent outcomes were included in the final model. Logistic regression was used for dichotomous outcomes (serum creatinine doubling, remission) and the Cox model for survival outcomes (renal death, transition into PLN). Predictors of outcome were analysed for each of the three treatment groups. Odds ratio (OR) was calculated to assess the risk of each variable.

Renal survival was estimated by the Kaplan–Meier method. Renal survival was calculated as the interval between the renal biopsy and the occurrence of end-stage renal failure. A subject who died prior to needing dialysis was considered as being censored out of the study. The assumption of proportional hazards was tested with a variable derived from the time to ESRD and was found to be acceptable.

Statistical analysis was performed with the SAS software (SAS Institute Inc.). All the tests were performed at a 0.05 significance level. Confidence intervals (CI) are at 95% level. Data are expressed as mean ± standard deviation.
Results

The incidence of LN classes Va and Vb was determined based on the renal biopsy register of one hospital (Pitié-Salpêtrière Hospital) of the seven involved in the study. Out of 260 patients diagnosed with LN undergoing a first renal biopsy between January 1976 and January 1997 in this hospital, 22 had a Va and Vb (8%).

Baseline characteristics

The cohort of 66 patients (mean age at the time of renal biopsy 31 ± 10 years) included 92% women. Forty-eight per cent were white, 47% were black, and 5% Asian. Cumulative ARA scores ranged from 2 to 8 (mean 4.5 ± 1.2). Eleven patients had cumulative ARA scores under 4 (16%) at baseline. For patients with cumulative ARA scores ≥ 4, mean follow-up time from the diagnosis of lupus to renal involvement was 22 ± 57 months during which 11 patients had pleural signs, 12 pericarditis, five central neurologic involvement, one valvulopathy, one myocarditis with cardiac failure, and four lung involvement (restrictive syndrome). Creatinine clearance was 97 ± 32 ml/min, serum albumin 25 ± 9 g/l, and proteinuria 4.0 ± 3.0 g/day. Serum albumin was in the nephrotic range in 64% and haematuria was present in 34% of the subjects. Sedimentation rate was 69 ± 41 mm and haemoglobin level 11.6 ± 2.2 g/dl. Immunological studies showed that nearly half of the patients had a decreased C3 complement (49%), 56% had significant level of serum antibodies to dsDNA (32/57), 41% had anti-Sm (18/44), 43% had anti-RNP (19/44), 25% had anti-SSA (11/44), 36% had anti-SSB (16/44), and 43% had antiphospholipid antibodies (18/41).

Analysis of the renal biopsies showed a mean glomerular count of 18 (± 10) with five renal biopsies having ≤ 5 glomeruli. Sclerotic glomeruli was observed on 35% of the biopsies (mean number of sclerotic glomeruli 1.3 ± 3.1 per biopsy, range 0–17). Hyaline deposits were observed in five biopsy specimens. Tubular atrophy was absent in 69%, mild in 22%, moderate in 7%, and marked in 2%. Interstitial inflammation was absent in 73%, mild in 20%, moderate in 5%, and marked in 2%. Fibrosis was absent in 66%, mild in 24%, and moderate in 10%. Pathological scores were: stage I in 19%, stage II in 69%, and stage III in 12%. Six patients had, in addition, predominant arteriolosclerosis and five had vascular lesions suggestive of the antiphospholipid syndrome.

After kidney biopsy, 18 patients were treated with corticosteroids ≥ 1 mg/kg/day, 32 patients with corticosteroids < 1 mg/kg/day, and 16 did not receive any treatment except antimalarials until the occurrence of another manifestation of lupus. Additional treatment included cyclophosphamide for three patients, azathioprine for four patients, and cyclosporin for one patient. Antimalarials were prescribed alone or with corticosteroids in 21 patients. The duration of treatments was highly variable. Three patients were being treated with angiotensin converting enzyme inhibitors at baseline and 23 during follow-up.

Outcome

Mean follow-up was 6.9 ± 0.2 years. Three patients died. In 30 patients (20%) serum creatinine doubled. Among them eight progressed to ESRD (12%). Renal survival at 5 and 10 years, calculated from Kaplan–Meier survival curves, was 97 ± 2 and 88 ± 6%, respectively, (Figure 1). Thirty-four patients (51%) experienced sustained renal remissions.

Twenty-nine patients underwent a second renal biopsy after 10.9 ± 1.1 years of follow-up (7.4 ± 1.2 years for patients who had a doubling of serum creatinine and 12.8 ± 1.4 years for the others, P < 0.03). Eleven patients whose serum creatinine doubled (11/13) and six patients who progressed to ESRD (6/8) underwent a second renal biopsy. Pathologic examination in 12 patients yielded a similar diagnosis compared with their initial biopsies (MLN class VA or VB), 14 patients had undergone transition into PLN and three patients had progressed to fibrosis. The probability of transformation into a PLN at 1, 5, and 10 years was 3 ± 2, 8 ± 4, and 35 ± 8%, respectively. Among patients who reached ESRD, four had a confirmed transition into PLN and their condition deteriorated despite immunosuppressive treatment, two had a confirmed progression to fibrosis, and two did not have repeat kidney biopsies. Each patient (n = 2) with progression to fibrosis had had three renal biopsies during follow-up without any proliferative lesions.

At the time of the second renal biopsy, serum creatinine was 182 ± 123 µmol/l for patients with transition to PLN, 291 ± 188 µmol/l for patients with class VI (NS), and 80 ± 18 µmol/l for patients with no pathologic evolution (P < 0.05). Complement C3 of the three patients with progression to fibrosis was normal, whereas it was diminished in 80% of patients with PLN or with VA-VB (P < 0.05).

Fifteen patients (23%) had a thrombotic event during follow-up. These events were: deep vein thrombosis (DVT) in seven patients (one sub- popliteal phlebitis, four femoral vein thrombosis, one retinal central vein thrombosis, one portal vein thrombosis) complicated by pulmonary embolism in two patients, pulmonary embolism without proven DVT in four patients, renal vein thrombosis in four patients complicated by pulmonary embolism in one and by inferior vena caval thrombosis in two patients. Thrombotic events were not significantly associated with nephrotic syndrome at baseline or with the presence of antiphospholipid antibodies. During follow-up, thrombosis was more frequent among patients with nephrotic syndrome (93% of patients with clinical thrombosis were nephrotic, P < 0.03).
Analysis of outcome

The development of ESRD was associated with marked hypoalbuminaemia at baseline (16 ± 5 g/l in patients with evolution to ESRD vs 27 ± 9 g/l in the others, \( P < 0.04 \)) and severe anaemia (Hgb 7.5 ± 1.3 g/dl in patients with evolution to ESRD vs 11.9 ± 2.1 g/dl in the others). Nephrotic syndrome at baseline was not associated with ESRD, but patients with a nephrotic syndrome at any time during follow-up reached ESRD more frequently (\( P < 0.04 \)). The haemoglobin level was the only predictor in the multivariate analysis. Each decrement of 1 g/dl of haemoglobin decreased renal survival by a factor of 0.61 (OR = 0.61 per unit decrease, CI [0.40–0.94]).

The predictors of doubling of serum creatinine are shown in Table 1. Thrombosis (\( P < 0.001 \)) and the occurrence of PLN (\( P < 0.001 \)) were the two predictors in the multivariate analysis.

The probability of transition into PLN was in direct relationship with the number of sclerotic glomeruli (OR = 1.13, CI [1.01–1.27], \( P < 0.04 \)) and with the occurrence of thrombosis during follow-up (OR = 3.03, CI [1.05–8.72], \( P < 0.04 \), multivariate analysis).

Remission was more frequent in patients without clinical thrombosis (\( P < 0.05 \)) or anti-SSA (\( P < 0.04 \), multivariate analysis).

Treatment with corticosteroids and ACE inhibitors was not associated with any prediction of outcome and did not affect renal survival in multivariate analysis.

Discussion

The former class V, which included proliferative subclasses Vc and Vd, accounted for 14% of LN in a
recent cohort of 1000 patients [4]. We calculated the frequency of VA-VB as 8% of LN. Subclasses VA-VB were already described separately in seven studies dedicated to MLN [5–10] and in two dedicated to all forms of LN [2,3].

The clinical presentation of patients with MLN is concordant with those studies and ours (Table 2). Patients often had nephrotic syndrome (50–69% of them) and normal renal function. Haematuria (25–50%) and hypertension were common. Complement was decreased in 6–59%. Some authors suggested that patients with pure MLN had a particular immunological status, with no or a low titre of anti-dsDNA, low DNA binding capacities, low or no precipitating DNA antibodies and no circulating immunological complexes [6,12]. Compared with a large cohort,
that included all patterns of disease expression in SLE [4], our cohort was marked by a rather low percentage of patients with anti-dsDNA. We also noticed a higher percentage of anti-Sm (41 vs 10% [4]), anti-RNP (43 vs 13%), and anti-SSB (36 vs 19%). This form of LN may also be present with no other clinical or serological manifestation of SLE. In our cohort, 11 patients had an ARA score or serological manifestation of SLE. In our cohort, 11 patients had an ARA score < 4, including renal signs, when MN was diagnosed. Several findings on immunofluorescence microscopy, as described by Jennette et al. [13], can suggest, however, underlying lupus rather than the idiopathic form of membranous nephropathy and SLE was confirmed for these patients by clinical evolution.

No major prognostic indicators of MLN could be identified in the literature. Bakir [2] identified thrombopenia < 40,000/mm³ as the sole prognostic feature. This finding probably is suggestive of superimposed thrombotic microangiopathy in patients with renal failure. Sloan [11] found serum creatinine as predictive of outcome but this association disappeared when subclasses Va to Vd were analysed separately. In our univariate analysis, a profound initial hypalbuminemia was a risk factor for ESRD and a sustained heavy proteinuria was a predictor of doubling of serum creatinine. As in idiopathic membranous nephropathy (IMN), proteinuria is of predictive value only when intense or when its duration is taken into account [14]. These factors, however, disappeared in multivariate analysis while factors more dependent on lupus activity persisted—haemoglobin level for ESRD and transition into PLN for doubling of serum creatinine.

A pathologic study was performed only by Radhakrishnan [9]: he found an almost significant association between the thickening of glomerular basement membrane and progression to renal failure. This association is partially confirmed by our study in which stage III is a predictor of doubling of serum creatinine but not of ESRD. Surprisingly, the presence of sclerotic glomeruli is associated with an increased probability of transition into PLN. Sclerotic glomeruli would be an indicator of a non-diagnosed proliferative flare-up of LN and are associated with an increased risk of a subsequent one.

Renal survival of our patients was 97 and 88% at 5 and 10 years, respectively. On a mean follow-up of 6.9 years, 12% of the patients reached ESRD. Renal death ranges from 4 to 22% in the literature (Table 2). Two different lesions may be responsible for the development of renal failure: an evolution mimicking the one of IMN with glomerulosclerosis and diffuse tubulointerstitial damages, and another one marked by transition into a PLN. In our cohort, only two patients progressed to ESRD without biopsy-proven PLN and two did not have a second renal biopsy. Concerning IMN, Hogan [15] reported a 35% incidence of ESRD after 10 years. Compared with this renal prognosis, the risk of ESRD, due only to glomerulofibrosis in MLN, seems lower.

The prognosis of MLN is more dependent on the risk of a transition into a PLN. The benefits of detecting this transition are reinforced by the fact that the prognosis of PLN is greatly modified by its treatment. We noted a high frequency of transition at 10 years, which was at least equal to 35%—considering that not all the patients underwent serial biopsies. Renal monitoring is particularly difficult for patients with persistent proteinuria. Among patients with renal failure, low complement C₃ seems to differentiate patients with transition to PLN from those with progression to fibrosis. In patients with normal renal function, no specific biological factor could be identified to detect a transition. During follow-up, thrombosis was identified as a predictor of PLN and of doubling of serum creatinine. Thrombosis occurs with an estimated frequency of 10–20% in SLE patients and seems even more frequent in patients with MLN. Pasquali [8] found an incidence of 23%, as in our study, and Radhakrishnan [9] of 28%. The high incidence of thrombosis suggests that anticoagulant therapy for patients with MLN should be used widely. Contrary to our study, Pasquali [8] found the presence of antiphospholipid antibodies to be a significant risk factor for thrombosis whereas, in our study the presence of a nephrotic syndrome diagnosed at any time during follow-up was the only risk factor for thrombosis. The high incidence of clinical thrombosis in this population probably is the result of multiple risk factors—mainly nephrotic syndrome, antiphospholipid antibodies, and SLE itself. Indeed, SLE with or without antiphospholipid antibodies proved to be responsible for a prothrombotic state. Positive correlation was observed between coagulation abnormalities and disease activity evaluated by anti-dsDNA levels and SLE Disease Activity Index (SLEDAI).

Therefore, thrombosis is due to an association of different thrombotic factors and its association with doubling of serum creatinine could have several explanations. In some patients, thrombosis may be associated with the persistent activity of the immunological disease that is responsible for renal damage. Less frequently, thrombosis may be associated with thrombi in glomerular capillaries, which explains the progression of renal failure. Different studies already have identified this latter type of course with hyaline thrombi made of fibrin and platelets in the glomeruli [16]. It seems a relatively rare event in SLE and not directly associated with disease activity but rather with antiphospholipid antibodies. Two patients in our cohort had antiphospholipid antibodies and this type of renal evolution. In one patient, renal biopsy at baseline revealed superimposed vascular lesions suggestive of the antiphospholipid syndrome. Thrombosis occurred during follow-up, and the patient progressed to ESRD without transition into PLN. Another patient with a history of repeated abortions presented with acute renal failure and convulsions at year 1 of follow-up. Renal biopsy disclosed transition into PLN and diffuse thrombotic microangiopathy.

Treatment of MLN remains controversial. Several clinical trials in patients with nephrotic MLN have
been already published. One trial [17] examined the efficacy of cyclophosphamide 2 mg/kg/day p.o. and prednisolone 0.8 mg/kg/day p.o. followed by azathioprine 2 mg/kg/day p.o. at 6 months. One patient out of six with classes Va and Vb achieved complete remission after 32 months of follow-up. Remission was less frequently observed in patients with Va-Vb than in the 22 class IV patients (77% in complete remission, P < 0.0001). The same authors [18] observed in 20 Va and Vb patients a 55% incidence of complete remission, 35% of partial remission and two non-responders (patients with lower baseline serum albumin compared with the complete responders). As in our study, profound hypoalbuminemia at baseline was a clear risk factor for adverse renal outcomes. Despite intensive immunosuppressive treatment, the rate of remission is comparable with that of nephrotic patients in our study (55% of remission).

Similarly, a retrospective study that included 15 nephrotic patients [19] class Va or Vb and four patients former class Vc compared the efficacy of corticosteroids alone (first group) vs a 6-month treatment alternating methylprednisolone and chlorambucil every other month (second group). In the second group, remission was more frequent and doubling of serum creatinine was less frequent compared with the first group. The only patient reaching ESRD was in the second group and had a confirmed transformation toward PLN 6 years after being diagnosed with MLN. In the first group, three patients developed renal failure at 2, 3, and 9 years but did not have renal biopsies after relapse. A transition into a PLN cannot be excluded in them. In our experience, this change more often is a transition into PLN than a progression to fibrosis.

The efficacy of adjunct immunosuppressive therapy with either cyclosporin or cyclophosphamide with alternate day prednisone was also observed in a recent randomized controlled trial of 41 patients [20]. Remission at 1 year was more frequent in the group with adjunct immunosuppressive therapy. Relapse tended to be more frequent with cyclosporin. Longer follow-up is needed for reliable conclusions.

The issue of cyclosporin was also addressed in a study including seven patients with pure MLN, two with class Vc, and one with Vd [21]. All patients except one were nephrotic and six were refractory to steroids or pulsed doses of cyclophosphamide plus corticosteroids (one patient). The mean duration of treatment was 21 months (range 6–43). At 12 months, proteinuria decreased from 6.5 to 2.2 g/day, serum albumin increased from 2.5 to 3.5 g/dl but serum creatinine increased from 8 to 9.5 mg/l. Seven patients underwent a second renal biopsy at 10 months. It is of note that in all cases, the activity index decreased but the chronicity index increased and the stage of MN increased from 2 to 3.

Regarding our experience and the results of these clinical trials, some conclusions about the treatment of MLN might be made. Profound hypoalbuminemia, anaemia at presentation, and persistent heavy proteinuria indicate patients with a high renal risk. The best therapeutic option might include corticosteroids with one adjunct immunosuppressive agent preferably not cyclosporin. During follow-up, the high rate of transition into a PLN reinforces the need of careful renal monitoring and justifies iterative renal biopsies in patients without remissions or followed by relapses.

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