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Clinicopathologic characteristic and prognosis in idiopathic membranous nephropathy patients with focal segmental sclerosis lesion

A retrospective observational study

Wenrong Cheng, MD, Lijun Sun, MM, Hongrui Dong, BS, Guoqin Wang, MD, Nan Ye, MM, Yanyan Wang, MD, Hong Cheng, MD*

Abstract

To explore the clinicopathological characteristics and prognosis of idiopathic membranous nephropathy (IMN) with focal segmental sclerosis lesions (FSL).

A total of 70 IMN patients with FSL (FSL+group) were enrolled in this study, and 140 patients were randomly selected by age and sex matching as disease controls (FSL-group). The clinical and renal histopathological data on renal biopsy and clinical data of patients regularly followed were collected. Serum anti-phospholipase A2 receptor (PLA2R) autoantibody, thrombospondin type-1 domain-containing 7A (7A) autoantibody, glomerular PLA2R and 7A expression, and IgG4 deposition were detected. First, the clinical and pathological significance of IMN combined with the FSL group was analyzed. Whether FSL is a risk factor for renal outcomes was further analyzed.

1. Compared with the FSL- group, patients in the FSL+ group had a significantly higher incidence of hypertension and a longer duration of hypertension as well as higher levels of systolic blood pressure, serum creatinine, serum triglycerides, serum cholesterol, 24-hour urinary protein excretion, and lower eGFR and urine osmotic pressure. Patients in the FSL+ group had an increased frequency of Churg stage III and more severe glomerulosclerosis and interstitial fibrosis. The remission rate was significantly lower in the FSL+ group than in the FSL- group (50.0% vs 75.9%, $P = .027$).
2. Multivariate Cox regression analysis showed that FSL (HR = 3.01, 95%CI = 1.07–8.52, $P = .038$) was an independent risk factor for progression of renal function deterioration, and FSL (HR = 3.25, 95%CI = 1.43–7.38, $P = .005$) and high levels of serum anti-PLA2R antibody (HR = 1.89, 95%CI = 1.27–2.82, $P = .002$) were independent risk factors for nonremission of IMN.

IMN patients who developed FSL had more severe clinical and pathological characteristics than those without FSL. FSL was an independent risk factor for poorer prognosis. When the appearance of FSL in IMN patients with a high level of serum anti-PLA2R antibody, the treatment needs to be more aggressive to promote remission and to delay the progression of renal function.

Abbreviations: ACEI = angiotensin-converting enzyme inhibitor, ARB = angiotensin receptor blockers, CI = confidence interval, CKD = chronic kidney disease, CR = complete remission, DM = diabetes mellitus, eGFR = estimated glomerular filtration rate, ELISA = enzyme-linked immunosorbent assay, ESRD = end-stage renal disease, FSL = focal segmental sclerosis lesions, HR = hazard ratio, HT = hypertension, IMN = idiopathic membranous nephropathy, IQR = interquartile range, KDIGO = Kidney Disease: Improving Global Outcomes, NS = nephrotic syndrome, PLA2R = M-type phospholipase A2 receptor, PR = partial remission, RU = relative units, SBP = systolic blood pressure, THSD 7A = thrombospondin type-1 domain-containing 7A.

Keywords: adhesion, focal segmental sclerosis, idiopathic membranous nephropathy, M-type phospholipase A2 receptor, prognosis

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The datasets generated during and/or analyzed during the current study are available from the corresponding author on reasonable request.

Division of Nephrology, Beijing Anzhen Hospital, Capital Medical University, Beijing, China.

* *Correspondence: Hong Cheng, Division of Nephrology, Beijing Anzhen Hospital, Capital Medical University, Beijing 100029, China (e-mail: drchengh@163.com).*

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1. Introduction

Primary glomerular disease (PGD) remains the leading cause of chronic kidney disease (CKD) in patients undergoing renal biopsies in China. Recently, with the changing spectrum of PGD in China, the proportion of idiopathic membranous nephropathy (IMN) has increased significantly, and it has become one of the most common PGDs, behind IgA nephropathy.[1,2] Although IMN seems to be a benign disease with a long-lasting natural course, with about one third of IMN patients having spontaneous remission, approximately 20% to 30% of patients progress to end-stage renal disease (ESRD) after 10 to 15 years without treatment.[3] Therefore, the risk factors affecting the prognosis of IMN are critical for clinical decisions.

IMN is one of the most common causes of nephrotic syndrome (NS) in adults, which is characterized by the formation of subepithelial immune complexes and thickening of the basement membrane. We found that focal segmental glomerulosclerosis and/or adhesion (named FSL) were present in pathological sections of some IMN patients, and the treatment response of these patients appeared to be poor during follow-up. Focal segmental sclerosis lesions in cases of IMN were first reported by Ehrenreich and Churg in 1977.[4] Damme et al[5] found that adhesions (approximately 51%) and focal sclerosis (approximately 33%) were frequently observed in IMN when serial sections were made. Therefore, in the recurrent study, the presence of FSL in patients with IMN was selected to be explored. The clinical and pathological characteristics of IMN patients with FSL were analyzed, and the effects of FSL on the prognosis and remission of proteinuria in IMN patients were further investigated and answered by the follow-up data, which can be obtained.

2. Materials and methods

2.1. Patients and samples

The study was approved by the Ethics Review Committee of Beijing Anzhen Hospital, Capital Medical University and was implemented in accordance with the Declaration of Helsinki. Informed consent was obtained for sampling the tissue and blood. A total of 806 patients with biopsy-proven IMN between January 2012 and January 2020 at our center were reviewed retrospectively. Patients with known secondary MN caused by systemic lupus erythematosus (SLE), hepatitis B/C virus, parvovirus B19, tumor, heavy metals and drug poisoning were excluded. Among the 806 IMN patients, 70 IMN patients with FSL were enrolled in this study, and 140 patients were randomly selected by age and sex matching from the remaining 736 patients as the disease control group.

In addition to the collection of duration from disease onset to renal biopsy and physical examination, the laboratory assessment included 24-hour urine protein quantity, urinary sediment analysis, urinary α1 microglobulin, urinary osmotic pressure, serum albumin, serum triglycerides, serum cholesterol, serum uric acid, serum creatinine and estimated glomerular filtration rate (calculated by eGFR-EPI formula)[6] were recorded at the time of biopsy. Hematuria was defined as a urinary red blood cell count greater than 3 per high-power field. Hypertension was defined as systolic blood pressure (SBP) exceeding 140 mm Hg and/or diastolic blood pressure exceeding 90 mm Hg, or currently receiving antihypertensive drugs.

2.2. Renal histopathology

All renal specimens were evaluated by light microscopy, direct immunofluorescence, and electron microscopy according to the standard procedure in our hospital.[7] MN was divided into 4 stages.[8] If 2 stages were noted at the same time, a relatively higher stage was selected. Each patient's renal biopsy included at least 8 glomeruli for histopathological evaluation. FSL was defined as focal segmental glomerulosclerosis and/or adhesion lesions (Fig. 1 A and B). The proportion of ischemic glomeruli = the number of glomeruli with ischemic sclerosis and shrinkage divided by the number of all glomeruli. The interstitial fibrosis was graded semiquantitatively from 0 to 3 (0, normal; 1, <25.0% of interstitium affected; 2, 25.0%–50.0% of interstitium affected; 3, >50.0% of interstitium affected).

2.3. Detection of serum anti-phospholipase A2 receptor (PLA2R) autoantibodies by enzyme-linked immunosorbent assay (ELISA) and serum thrombospondin type-1 domain-containing 7A (7A) autoantibodies by indirect immunofluorescence.

Serum samples were obtained on the day of the renal biopsy. After collection, the serum was immediately centrifuged and stored at −80°C until use. The level of serum anti-PLA2R antibody was detected using a commercially available ELISA kit, which was purchased from EUROIMMUN Medizinische Labordiagnostika AG. The results were considered positive at ≥20 relative units (RU)/ml. The level of anti-7A antibody was detected using an indirect immunofluorescence kit from the same company.

2.4. Detection of glomerular PLA2R and 7A expression by immunohistochemical staining and IgG subclasses deposition by direct immunofluorescence

The glomerular PLA2R, 7A, and IgG subclasses were detected by a stable procedure, which has been described by Dong et al.[7] Positivity of glomerular PLA2R and 7A expression was defined as granular or linear diffuse staining on glomeruli (Fig. 1C and D).

2.5. Follow-up records

The date of renal biopsy was taken as the starting point, and the last follow-up time or the date of loss of visit or development of the primary renal end point event was taken as the end point. The follow-up time was at least 6 months. Patients received treatments according to the Kidney Disease: Improving Global Outcomes (KDIGO) guidelines for glomerulonephritis.[9] During follow-up, complete remission (CR) was defined as urinary protein excretion <.3 g/d. Partial remission (PR) was defined as urinary protein excretion <3.5 g/d and at least 50% reduction verus baseline. Relapse was defined as proteinuria increase to ≥3.5 g/d in patients with previous CR or PR. The primary end point was defined as a 25% decline in baseline eGFR or ESRD (defined as eGFR <15 ml/minute/1.73 m^2 or starting renal replacement therapy). Severe complications were defined as severe infection, pulmonary embolism, deep vein thrombosis (such as lower extremity vein, renal vein, or inferior vena cava thrombosis), and cerebral infarction.

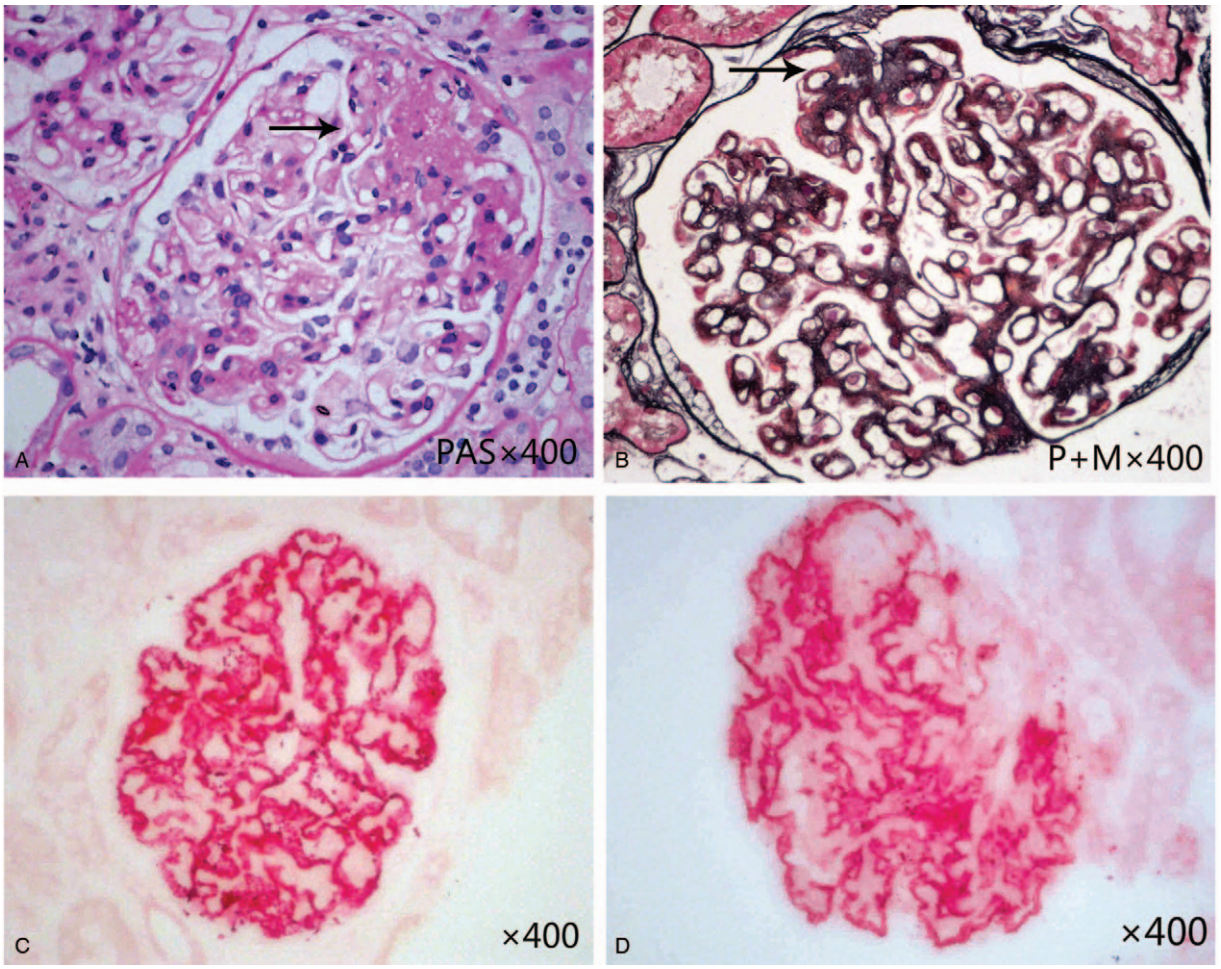

Figure 1. A: the black arrow indicates focal segmental glomerulosclerosis; B: the black arrow indicates an adhesion lesion; C: positivity of glomerular expression of PLA2R; D: positivity of glomerular expression of 7A. (PAS = periodic acid-Schiff stain, P+M = periodic acid-silver methenamine and Masson stain).

2.6. Statistical analysis

SPSS 25.0 statistical software was used for data analysis. Quantitative variables with normal distributions were expressed as $x \pm s$ and compared by t tests, and data with abnormal distributions were expressed as median and interquartile range (IQR), compared by nonparametric tests. The qualitative variables were compared using the χ^2 test or Fisher exact test. The Cox proportional hazards model was used to identify candidate and independent predictors. P value $<.05$ was considered statistically significant.

3. Results

3.1. Baseline demographic and clinical characteristics of IMN patients

A total of 210 IMN patients were enrolled in this study. Among them, 70 IMN patients were combined with FSL (FSL+ group), and 140 were treated with IMN without FSL (FSL- group). A comparison of demographic and clinical parameters between these 2 groups is presented in Table 1. Compared with patients in the FSL- group, those in the FSL+ group had a higher frequency of primary hypertension history, a longer duration of hypertension, and a higher level of SBP ($P < .05$). The proportion of NS in the FSL+ group was higher than that in the FSL-group ($P < .05$). The levels of serum creatinine, serum triglycerides, serum cholesterol, and 24-hour urinary protein excretion in the FSL+ group were higher than those in the FSL-group ($P < .05$). The levels of urinary osmotic pressure and eGFR in the FSL+ group were lower than those in the FSL-group ($P < .05$). There was no significant difference between the 2 groups in the proportion, duration time of type 2 diabetes mellitus at presentation, level of serum albumin and microscopic hematuria frequency ($P > .05$).

3.2. Baseline pathological parameters

All enrolled patients mainly presented with Churg stage I and II IMN, without Churg stage IV. A total of 21 (30.0%) patients in the FSL+ group presented with Churg stage III, while 7 patients (5.0%) presented with stage III in the FSL-group ($P < .05$). Patients in the FSL+ group presented with more severe glomerulosclerosis and interstitial fibrosis ($P < .05$). There was no significant difference in the deposition of IgG and IgM between the 2 groups. IgG4 was the predominant IgG subclass in the glomeruli of both groups, and there was no statistically significant difference. All the findings are shown in Table 2.

Table 1

Comparison of clinical parameters of IMN patients among 2 groups at baseline.

	IMN (n = 210)	FSL+ (n = 70)	FSL- (n = 140)	*P* value
Gender (male/female)	139/71	46/24	93/47	.918
Age (year)	47.9 ± 13.8	48.7 ± 13.2	47.4 ± 14.1	.525
Duration of illness (month)	3.0 (1.0,8.3)	4.0 (1.0,9.0)	2.0 (1.0,7.0)	.112
HT (n,%)	102 (48.6)	45 (64.3)	57 (40.7)	.001
Duration of HT (month)	0 (0,60)	12 (0,87)	0 (0,48)	.001
DM (n,%)	17 (8.1)	7 (10.0)	10 (7.1)	.474
Duration of DM (month)	0 (0,0)	0 (0,0)	0 (0,0)	.716
Waistline (cm)	88.8 ± 10.4	90.3 ± 9.9	88.2 ± 10.5	.178
Body mass index (kg/ m^2)	25.2 ± 3.8	25.9 ± 3.9	24.9 ± 3.8	.106
SBP (mm Hg)	133 ± 21	137 ± 18	130 ± 22	.038
DBP (mm Hg)	84 ± 12	86 ± 12	83 ± 12	.140
Hematuria (n,%)	112 (53.3)	36 (51.4)	76 (54.3)	.696
24-h urinary protein (g/d)	5.5 (3.6,9.5)	7.1 (4.5,10.5)	5.2 (2.9,7.7)	.001
Serum albumin (g/L)	25.1 ± 7.1	24.0 ± 6.9	25.7 ± 7.2	.116
NS (n,%)	164 (78.1)	61 (87.1)	103 (73.6)	.025
Serum creatinine (μmol/L)	68.8 (58.0,88.4)	78.2 (59.2,96.6)	67.0 (58.0,83.0)	.018
eGFR-EPI (ml/min/1.73 m^2)	97.0 ± 23.8	89.5 ± 27.5	100.8 ± 20.9	.003
eGFR < 60 ml/min (n,%)	16 (7.6)	12 (17.1)	4 (2.9)	<0.001
Urinary osmotic pressure (mOsmol/kg)	639.8 ± 188.5	579.2 ± 175.8	667.1 ± 188.4	.008
Urinary α1 microglobulin (mg/L)	42.1 (18.9,76.6)	46.4 (20.8,103.8)	40.3 (18.1,66.1)	.239
Serum triglyceride (mmol/L)	2.1 (1.5,3.3)	2.3 (1.8,3.8)	2.0 (1.4,3.1)	.010
Serum cholesterol (mmol/L)	7.3 (6.0,9.1)	7.7 (6.7,9.3)	7.1 (5.7,9.0)	.029
Serum uric acid (μmol/L)	370.7 ± 93.5	371.8 ± 94.6	370.1 ± 93.3	.900

P < .05 between Group FSL+ and FSL−.

DBP = diastolic blood pressure, DM = diabetes mellitus, eGFR = estimated glomerular filtration rate, FSL = IMN patients combined with focal segmental sclerosis lesions, HT = hypertension, IMN = primary membranous nephropathy, NS = nephrotic syndrome, SBP = systolic blood pressure.

3.3. Baseline serum autoantibodies and glomerular expression of PLA2R and 7A

We examined circulating anti-PLA2R autoantibodies and glomerular PLA2R expression in all enrolled patients. The levels of anti-PLA2R autoantibodies were comparable between the FSL + and FLS- groups. Granular or linear staining of PLA2R in the glomeruli was observed in 95.7% of patients in the FSL+ group, which was similar to the patients in the FSL- group (95%). There were 10 PLA2R-negtive patients—3 patients in the FSL+ group, and 7 patients in the FSL- group. Only 6 of the 10 patients whose serum and renal specimens were sufficient to perform 7A related detection, circulating anti-7A antibodies and glomerular 7A expression were all negative. All the findings are shown in Table 3.

3.4. Treatment and therapeutic response during follow-up

A total of 76 patients were regularly followed in our center, and the mean follow-up time was 16.5 months (range, 6–89 months). The total remission rate (CR and PR) was 68.4%, with approximately 25% of patients relapsing. Patients received treatments according to the KDIGO guidelines for IMN.[9] The patients received specific treatments, including glucocorticoids plus cyclosporine A/cyclophosphamide, cyclosporine A only, and angiotensin-converting enzyme inhibitors (ACEI)/angiotensin receptor blockers (ARB) only.

Table 2

Comparison of pathological parameters of IMN patients among 2 groups at baseline.

	IMN(n = 210)	FSL+(n = 70)	FSL-(n = 140)	*P* value
IMN stage				<.001
I+II(n, %)	182 (86.7)	49 (70.0)	133 (95.0)	
III(n, %)	28 (13.3)	21 (30.0)	7 (5.0)	
Interstitial fibrosis				<.001
Grade 0 (n, %)	39 (18.6)	3 (4.3)	36 (25.8)	
Grade 1(n, %)	135 (64.3)	48 (68.6)	87 (62.1)	
Grade 2(n, %)	27 (12.9)	13 (18.6)	14 (10.0)	
Grade 3(n, %)	9 (4.3)	6 (8.6)	3 (2.1)	
Proportion of ischemic glomeruli (%)	5.4 (0,14.4)	8.0 (0,22.2)	4.2 (0,12.5)	.007
Glomeruli IgG deposit (n,%)	210 (100.0)	70 (100.0)	140 (100.0)	–
Glomeruli IgG4 deposit predominantly (n,%)	186 (89.0)	60 (87.0)	126 (90.0)	.509
Glomeruli IgM deposit (n,%)	33 (15.7)	11 (15.7)	12 (8.6)	.118

P < .05 between Group FSL+ and FSL−.

FSL = IMN patients combined with focal segmental sclerosis lesions, IMN = primary membranous nephropathy.

Table 3

Comparison of serum anti-PLA2R and anti-7A autoantibodies and glomerular PLA2R and 7A expression of IMN patients among 2 groups at baseline.

	IMN (n = 210)	FSL+ (n = 70)	FSL- (n = 140)	*P* value
Glomerular PLA2R expression (n,%)	200 (95.2)	67 (95.7)	133 (95.0)	1.000
Serum anti-PLA2R antibody (RU/ml)	44.8 (11.5,102.9)	46.5 (12.3,136.7)	42.4 (8.1,91.2)	.200
Glomerular THSD7A Expression (n,%)	0/6 (0)	0/1 (0)	0/5 (0)	–
Serum anti-7A antibody (n,%)	0/6 (0)	0/1 (0)	0/5 (0)	–

$P<.05$ between Group FSL+ and FSL−.

FSL = IMN patients combined with focal segmental sclerosis lesions, IMN = primary membranous nephropathy, PLA2R = M-type phospholipase A2receptor, THSD7A = thrombospondin Type-1 Domain-Containing 7A.

There were no differences in treatment between the 2 groups ($P > .05$). In this cohort, 22 patients were in the FSL+ group, and the remaining 54 patients were in the FSL- group. The total remission rate (CR and PR) in the FSL+ group was 50.0%, which was significantly lower than that in the FSL- group (75.9%) ($P < .05$). The numbers of CR and PR patients were 4 and 7 in the FSL+ group and 20 and 21 in the FSL- group, respectively. There were no significant differences in remission duration and relapse rate between the 2 groups ($P > .05$) (Table 4). We performed a subgroup analysis based on different treatments. When patients received cyclosporine A alone, the total remission rate of the FSL+ group was significantly lower than that of the FSL- group($P < .05$), while there was no difference in relapse ($P > .05$). When patients received other treatments, such as glucocorticoids plus cyclosporine A/cyclophosphamide, and ACEI/ARB only, the total remission and relapse rates were similar between the 2 groups (Table 4). In addition, the levels of serum anti-PLA2R autoantibodies in the remission patients (CR and PR) were significantly lower than those in the nonremission patients ($P < .05$), while there was no difference in glomerular PLA2R deposits (Table 5).

3.5. Clinical prognosis analysis

At the end of follow-up, 19 patients (25%) experienced a 25% decline in eGFR (including progression to ESRD), of which 1 (1.3%) progressed to ESRD. Among them, there were 6 cases in the FSL+ group and 3 cases in the FSL- group. During follow-up, severe complications were observed in 4 patients—1 patient in the FSL+ group experienced pulmonary embolism and renal vein thrombosis and 3 patients in the FSL- group, including spleen infarction in 1 patient, deep vein thrombosis of the lower extremity in 1 patient, and acute diffuse peritonitis in 1 patient.

Univariate analyses were used to explore potential associations of clinical outcomes (25% decline in eGFR, including ESRD) with a series of variables (age, male sex, 24-hour urinary protein excretion, eGFR, SBP, and FSL). Age, male sex, SBP, and FSL were identified as potential predictors of renal outcomes. Then, Cox proportional hazards models were used. After adjustment, FSL was significantly independently associated with a 25% decline in eGFR, as shown in Table 6. Additionally, a Cox proportional hazard model was used to analyze age, male sex, 24-hour urinary protein excretion, eGFR, SBP, FSL, serum anti-PLA2R autoantibody, and treatment option (cyclosporine A only). FSL and high levels of serum anti-PLA2R autoantibody were found to be independent risk factors for nonremission, as shown in Table 7.

Table 4

Treatment and therapeutic response of 76 patients during follow-up.

	FSL+(n = 22)	FSL-(n = 54)	*P* value
CR&PR (n,%)	11 (50.0)	41 (75.9)	.027
Duration of CR or PR (month)	8.0 (6.0,16.0)	9.0 (6.3,18.0)	.583
Relapse (n,%)	3 (27.3)	10 (24.4)	.846
Treatment			
ciclosporin only (n,%)	11 (50.0)	31 (57.4)	.556
CR&PR (n,%)	3 (27.3)	22 (71.0)	.029
relapse (n,%)	0 (0)	6 (27.3%)	.283
ciclosporin&semis glucocorticoid (n,%)	9 (40.9)	11 (20.4)	.065
CR&PR (n,%)	6 (66.7)	9 (81.8)	.617
relapse (n,%)	2 (33.3)	3 (33.3)	1.000
other (n,%)*	2 (9.1)	12 (22.2)	.327
CR&PR (n,%)	2 (100)	10 (83.3)	1.000
relapse (n,%)	1 (50.0)	1 (10.0)	.318

* Other: including glucocorticoid plus cyclophosphamide or use of angiotensin-converting enzyme inhibitor/angiotensin type 1 receptor blockers only.

CR = complete remission, FSL = IMN patients combined with focal segmental sclerosis lesions, PR = partial remission.

4. Discussion

Recently, increasing amount of attention has been given to the pathomorphological characteristics of IMN, especially FSL. In fact, FSL is not uncommon in IMN, and the incidence is between 10 and 41.5%,[10–14] which is in contrast to our incidence of 8.5%. The clinical phenotype and renal pathological characteristics of patients with combined IMN and FSL have been reported in many previous studies since 1977. Li et al[10] found that IMN patients with FSL had a longer course of disease, higher levels of blood pressure, and higher serum creatinine. Dumoulin et al[15] suggested that IMN patients with FSL had a higher frequency and increased degree of hypertension, but no differences in proteinuria and serum creatinine were found. Gu et al[14] showed that

Table 5

Comparison between baseline serum anti-PLA2R antibodies and therapeutic response during follow-up of 76 patients.

	CR&PR (n = 52)	No Remission (n = 24)	*P* value
Glomerular PLA2R expression (n,%)	50 (96.2)	24 (100)	1.000
Serum anti-PLA2R antibody (RU/ml)	45.0 (16.2,82.7)	124.5 (32.1,338.8)	.010

CR = complete remission, PLA2R = M-type phospholipase A2 receptor, PR = partial remission, RU = relative units.

Table 6

Multivariate analysis of risk factors for progression to primary outcome–Cox proportional hazards model.

	Univariate analysis			Multivariate analysis		
Variables	**HR**	**95%CI**	***P* value**	**HR**	**95%CI**	***P* value**
Age (year)	1.02	0.99–1.05	.150	1.02	0.99–1.05	.145
Gender(male)	2.33	0.88–6.18	.087	2.17	0.97–7.06	.057
24-h urinary protein (g/d)	1.05	0.94–1.17	.402	–	–	–
eGFR (ml/min/1.73 m^2)	0.99	0.97–1.01	.312	–	–	–
SBP (mm Hg)	1.02	0.99–1.04	.198	1.01	1.00–1.03	.708
FSL	2.68	0.97–7.37	.057	3.01	1.07–8.52	.038

CI = confidence interval, eGFR = estimated glomerular filtration rate, FSL = IMN patients combined with focal segmental sclerosis lesions, HR = hazard ratio, SBP = systolic blood pressure.

hypertension incidence, abnormal serum creatinine levels, microscopic hematuria incidence, and 24-hour urinary protein excretion were higher in the FSL+ group than in the FSL- group. On histopathological examinations, previous studies have shown that IMN patients with FSL presented with later stages of MN and presented with more serious pathological manifestations, such as more severe glomerulosclerosis, mesangial hyperplasia, endothelial hyperplasia, interstitial fibrosis, interstitial infiltration, and tubular atrophy.[10–12,14–16] Our analysis shows that IMN patients with FSL had a high incidence of hypertension (64.3%), and had a longer duration of hypertension. In the FSL+ group, the level of systolic blood pressure, serum creatinine, serum triglycerides, serum cholesterol, and 24-hour urinary protein excretion were higher, while the level of eGFR was lower. The IMN patients with FSL in our study presented with later Churg stage (without Churg stage IV), and an increased frequency of Churg stage III was observed in the FSL+ group compared to that in the FSL- group. Renal pathological damage, including glomerulosclerosis and interstitial fibrosis, was more severe in the FSL+ group. In summary, IMN patients with FSL had more severe clinical and pathological characteristics than those without FSL.

IMN is an autoimmune glomerulopathy, and the target antigen has been identified as PLA2R[17] and 7A[18] in approximately 70% and 1% to 5% of IMN, respectively. We further analyzed the level of serum anti-PLA2R autoantibody and glomerular PLA2R deposits between these 2 groups. There were no differences in these 2 markers between the 2 groups. There were 10 PLA2R-negtive patients, only 6 with sufficient serum and renal specimens to perform circulating anti-7A autoantibodies and glomerular 7A expression detection. All were negative. Novel antigens, such as neural epidermal growth factor-like 1protein (NELL-1), may be the antigen responsible for IMN.[19]

Clinical and pathological markers, which can predict renal progression or remission, are important for clinicians to make decisions. Several predictive markers of an unfavorable course at presentation have been determined, including age, male sex, heavy proteinuria, renal insufficiency, hypertension, interstitial fibrosis, arteriosclerosis, and serum anti-PLA2R autoantibody.[20–23] However, debate continues about whether the existence of FSL contributes to more severe histopathological damage and worse prognosis in IMN. Previous studies have shown that the primary endpoints were kidney function insufficiency or 25%/30%/50% decline in initial eGFR or doubling of baseline creatinine or ESRD. Conclusions of prognosis were controversial with short-term or long-term follow-up and need to be further studied. Li,[10] Heeringa,[12] Troyanov,[24] et al suggested that the FSL was not an independent risk factor in patients with IMN. Meanwhile, Wakai and Magil,[11] Dumoulin,[15] Chen,[13] et al showed that FSL was an independent risk factor. Kanigicherla et al[25] suggested that high levels of anti-PLA2R autoantibody at presentation were linked with a higher risk of doubling of baseline creatinine during 5 years follow-up by survival analysis. In our enrolled cohort, the mean follow-up time was 16.5 months, and a 25% decline in initial eGFR (including ESRD) was the primary end point. FSL was an independent risk factor for renal function progression in the Cox proportional hazards model. In addition, our study

Table 7

Multivariate analysis of risk factors for therapeutic response–Cox proportional hazards model.

	Univariate analysis			Multivariate analysis		
Variables	**HR**	**95%CI**	***P* value**	**HR**	**95%CI**	***P* value**
Age (year)	1.00	0.97–1.03	.826	1.00	0.96–1.03	.811
Gender (male)	2.07	0.91–4.71	.081	1.70	0.70–4.10	.240
24-h urinary protein (g/d)	1.05	0.98–1.13	.194	1.04	0.95–1.15	.372
eGFR (ml/min/1.73 m^2)	1.00	0.98–1.02	.843	–	–	–
SBP (mm Hg)	1.01	0.99–1.03	.475	–	–	–
Serum anti-PLA2R antibody*	1.89	1.25–2.76	.002	1.89	1.27–2.82	.002
Treatment option† (ciclosporin only)	1.74	0.72–4.21	.216	1.47	0.60–3.63	.400
FSL	3.06	1.36–6.90	.007	3.25	1.43–7.38	.005

* The serum anti-PLA2R antibody was graded according to the titer quartile spacing.

† Treatment option was ciclosporin only VS other treatment plans (such as ciclosporin&semis glucocorticoid, glucocorticoid combined with cyclophosphamide, angiotensin-converting enzyme inhibitor/angiotensin receptor blockers only).

CI = confidence interval, eGFR = estimated glomerular filtration rate, FSL = IMN patients combined with focal segmental sclerosis lesions, HR = hazard ratio, PLA2R = M-type phospholipase A2 receptor, SBP = systolic blood pressure.

found that the total remission rate was lower in IMN patients with FSL, and the level of serum anti-PLA2R antibody was significantly higher in patients with non-remission IMN, and further analysis showed that FSL and high levels of serum anti-PLA2R autoantibody were independent risk factors for non-remission by the Cox proportional hazards model. However, decreased eGFR and heavy proteinuria showed no effect in predicting kidney outcomes or the treatment responses of patients with IMN.

In addition, the total remission rate of the FSL+ group was significantly lower when patients received cyclosporine A alone. We speculated that it may be more appropriate to choose glucocorticoids plus cyclosporine A or glucocorticoids plus cyclophosphamide A in IMN patients with FSL. To what extent did FSL contribute to nonremission and whether more aggressive treatments should be considered for these patients need to be answered by randomized clinical trials.

To date, the existence of FSL in pathological changes of IMN may be important to predict outcome; however, the potential etiology and mechanism of FSL in IMN remains unclear. First, hemodynamic changes, such as systemic hypertension and increased glomerular capillary pressure, hyperperfusion, and hyperfiltration, which would alter glomerular permeability, and cause an abolition of the selectivity barriers, resulted in a nonselective leak of plasma-like material and further development of focal glomerulosclerosis.[5,11] In the current study, hypertension was found in more than half of IMN patients with FSL. Second, some studies have suggested that the subepithelial immune complex deposits contribute to the detachment of podocytes from the glomerular basement membrane through $\alpha 3\beta 1$ integrins and the redistribution of podocyte adhesion molecules, which may be one of the causes for the formation of FSL in MN.[14] However, this needs further investigation.

In conclusion, IMN patients with FSL were more likely to have hypertension, had a lower baseline eGFR, heavier proteinuria, and more severe tubulointerstitial and glomerulosclerosis damage. FSL was an independent risk factor for predicting poor outcome. When FSL appears in patients with IMN, meanwhile with high levels of serum anti-PLA2R autoantibody, the treatment needs to be more aggressive to promote remission and to delay the progression of renal function.

Of course, there are some limitations to our study. First, this was a single-center retrospective clinical study. Second, the observation time was short, and the follow-up patients were few. Therefore, further large-scale clinical trials and mechanistic research are needed to further confirm these results.

Author contributions

Conceptualization: Wenrong Cheng, Hong Cheng.
Data curation: Wenrong Cheng, Guoqin Wang, Nan Ye, Yanyan Wang.
Formal analysis: Wenrong Cheng.
Investigation: Wenrong Cheng.
Methodology: Lijun Sun, Hongrui Dong.
Project administration: Hong Cheng.
Resources: Yanyan Wang.
Supervision: Guoqin Wang, Hong Cheng.
Validation: Hongrui Dong.
Writing – original draft: Wenrong Cheng.
Writing – review & editing: Wenrong Cheng, Hong Cheng.

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