Urinary Albumin Levels are Independently Associated with Renal Lesion Severity in Patients with Lupus Nephritis and Little or No Proteinuria

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Background: Systemic lupus erythematosus (SLE) leads to renal lesions, which may be clinically silent in patients with little or no proteinuria. Early detection of these lesions may improve prognosis, but early markers are controversial. This study aimed to determine renal marker proteins associated with renal lesion severity in patients with lupus nephropathy (LN) and little or no proteinuria.

Material/Methods: Patients with LN and little or no proteinuria (<0.5 g/24 hours) (n=187) that underwent kidney biopsy were grouped according to: low severity (Class I or II; n=116) versus high severity (Class III, IV, or V; n=71). Disease status was determined according to the SLE disease activity index (SLEDAI). Renal marker proteins (serum β2-macroglobulin, urinary β2-macroglobulin, albumin, IgG, and α1-macroglobulin) were measured using radioimmunoassay.

Results: Compared with the low severity group, patients in the high severity group had higher urinary albumin (11.60±8.94 versus 7.08±10.07 μg/mL, p=0.008) and urinary IgG (13.21±9.35 versus 8.74±8.90 μg/mL, p=0.007) levels. Multivariate conditional logistic regression analysis showed that urinary albumin (odds ratio (OR)=1.417, 95% confidence interval (95% CI): 1.145–1.895, p=0.001) and SLEDAI (OR=2.004, 95% CI: 1.264–3.178, p=0.003) were independently associated with severe renal lesions in these patients. Using an optimal cutoff point of urinary albumin of 7.53 μg/mL resulted in 67% sensitivity and 82% specificity for the detection of high severity renal lesions.

Conclusions: Urinary albumin levels and SLEDAI were independently associated with histological severity of renal lesions in patients with LN and little or no proteinuria. These parameters could be used to help select patients for renal biopsy.

MeSH Keywords: Acute Kidney Injury • Lupus Nephritis • Proteinuria

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Background

Lupus nephritis (LN) occurs in 40–70% of patients with systemic lupus erythematosus (SLE) and is among the most serious complications of SLE [1]. The incidence of LN is higher in Asians (55%) compared with Caucasians (14%) [1,2]. In China, LN is the most common renal disease and accounts for 54.3% of all secondary glomerular diseases [3]. The manifestations of LN range from asymptomatic urinary findings to nephrotic syndrome and progressive renal impairment. The International Society of Nephrology and the Renal Pathology Society (ISN/RPS) have indicated that the various LN classes exhibit different natural history and clinical patterns [4]. ISN/RPS Classes I and II usually display low activity and chronicity index, and have an indolent course. On the other hand, Classes III, IV, and V are progressive types of LN [5]. For Class IV, the 5-year probability of developing end-stage renal disease (ESRD) is as high as 70% [6]. Early and accurate detection of kidney involvement is needed to improve the prognosis of patients with LN.

Generally, urinalysis is the most effective method for detecting and monitoring LN disease severity by assessing the dominant feature of LN, i.e., proteinuria [7], but previous studies casted doubts on the ability of urinary findings to predict the underlying presence and severity of nephritis, at least at the early stage [8–12]. In addition, some patients may have significant kidney damage and still be asymptomatic, at least based on conventional urine disease markers [13,14]. Renal biopsy is required for diagnosing LN and planning therapy, but this approach is invasive and a biopsy is ethically acceptable only for patients with at least a suspicion of LN [6]. Furthermore, studies have suggested that significant kidney damage (Class III, IV, or V) may be present in many patients with SLE, but without any sign of renal involvement, i.e., in the presence of little or no proteinuria (<0.5 g/24 hours) [5,15,16].

In asymptomatic patients, the total amount of urine protein may be small, but some specific proteins could predict or indicate kidney damage. Indeed, proteins of high molecular weight such as IgG (150 kD) are excreted in large quantities when permselectivity of the glomerular capillary wall is severely disrupted [17,18]. Therefore, these proteins could be markers of the severity of glomerular damage. Proteins of low molecular weight, such as α1-microglobulin (α1-MG) (31.8 kD) or β2-microglobulin (β2-MG) (11.8 kD), are excreted in large quantities when proximal tubular cells lose their capacity to reabsorb them; therefore, these proteins reflect intrinsic tubular injury [19]. Studies have shown that serum and urinary levels of β2-MG were associated with renal involvement in SLE [20,21], as well as urinary albumin (UAL) [22], IgG, and α1-MG [23]. Physiologically, α1-MG is involved in the defense against oxidative tissue damage [24], while β2-MG is a component of the MHC class I [25]. Nevertheless, their exact predictive values

are controversial and additional studies are still necessary to address their value for the early detection of LN.

Therefore, the present study aimed to find renal marker proteins that could be associated with renal histopathological findings, in order to provide bases for early diagnosis of LN in patients with little or no proteinuria.

Material and Methods

Patients

This study was approved by the ethics committee of the Xijing Hospital, Fourth Military Medical University (Xi'an, China) (No. 20110303-6). Written informed consent was obtained from each participant.

The study included 187 patients with LN and little or no proteinuria (as based on previous studies, <0.5 g/24 hours [6,12]) that underwent kidney biopsy at the Department of Clinical Immunology of Xijing Hospital between April 2005 and November 2010. Inclusion criteria were: 1) all patients were confirmed with LN by biopsy and fulfilled the 1982 American College of Rheumatology (ACR) revised criteria for SLE [26]; and 2) little or no proteinuria (<0.5 g/24 hours). Patients with renal failure and serum creatinine levels >2 mg/dL were excluded.

Indications for renal biopsy include patients with low or no proteinuria (<0.5 g/24 hours) and with or without hematuria (urinary red blood cell (RBC) count >25/μL as analyzed by flow cytometry) or pyuria (urinary white blood cell (WBC) count >25/μL), or accompanied by: 1) abnormal increase of renal marker proteins (serum β2-MG >3 μg/mL, urine β2-MG >160 ng/mL, urinary albumin >6 μg/mL, urinary IgG >5.5 μg/mL, or urinary α1-MG >5 μg/mL); 2) hypocomplementemia (C3 <79 mg/dL and/or C4 <16 mg/dL), and/or 3) positive anti-double stranded DNA (dsDNA) and/or positive anti-nucleosome antibodies [27,28].

Histological renal tissue subtypes (Classes I-VI) were determined according to the 2003 ISN/RPS LN classification [4]. LN patients with little or no proteinuria (n=187) were divided into two groups: low severity (Classes I and II LN) (group 1) and high severity (Classes III, IV, and V) (group 2), according to a previous report [5].

The demographic characteristics and clinical manifestations of the 187 patients with LN and little or no proteinuria are presented in Table 1. There were 172 (92.0%) women, for a female-to-male ratio of 11.5: 1, and 184 patients (98.4%) were of Chinese Han ethnicity. The mean age was 31.7±11.2 years, the mean age at disease onset was 29.6±10.8 years, and the mean disease duration was 44.0±52.2 months. Among all patients,

Table 1. General characteristics and clinical manifestations in LN patients with little or no proteinuria according to histologic classification of kidney biopsies.

Variables	Low severity (Classes I and II) (n=116)	High severity (Classes III, IV, and V) (n=71)	p
Female, n (%)	110 (94.8)	62 (87.3)	0.067
Age at onset (years)	30.9±10.6	29.3±9.9	0.402
Disease duration (months)	42.3±50.4	50.6±60.1	0.410
Time between symptom onset and SLE diagnosis (months)	32.8±48.6	24.6±41.6	0.331
Time between LN onset and LN diagnosis (months)	4.7±13.5	8.8±17.1	0.148
Time between LN onset and biopsy (months)	8.0±23.6	20.5±33.2	0.022*
Fever, n (%)	54 (46.6)	44 (62.0)	0.040*
Malar rash, n (%)	50 (43.1)	37 (52.1)	0.231
Photosensitivity, n (%)	34 (29.3)	14 (19.7)	0.145
Oral ulcers, n (%)	21 (18.1)	22 (31.0)	0.042*
Alopecia, n (%)	70 (60.3)	44 (62.0)	0.825
Raynaud phenomenon, n (%)	47 (40.5)	21 (29.6)	0.131
Arthritis, n (%)	89 (76.7)	56 (78.9)	0.733
Serositis, n (%)	23 (19.8)	16 (22.5)	0.658
Myositis, n (%)	18 (15.5)	9 (12.7)	0.592
Neurological disorder, n (%)	8 (6.9)	8 (11.3)	0.300
Leucopenia, n (%)	56 (48.3)	31 (43.7)	0.539
Anemia, n (%)	39 (33.6)	29 (40.8)	0.319
Thrombocytopenia, n (%)	20 (17.2)	26 (36.6)	0.003**
Hypertension, n (%)	12 (10.3)	20 (28.2)	0.002**
SLEDAI	7.7±6.5	12.5±6.6	<0.001***
C3 (mg/dl)	72.3±30.9	58.0±28.3	0.003
Low C3, n (%)	59 (50.9)	59 (83.1)	<0.001***
C4 (mg/dl)	14.5±12.1	11.6±8.9	0.150
Low C4, n (%)	62 (53.4)	51 (71.8)	0.014*
Proteinuria			
<0.3 g/24 h, n (%)	98 (84.5)	52 (73.2)	0.088
0.3–0.5 g/24 h, n (%)	18 (15.5)	19 (26.8)	
Hematuria (%)	15 (12.9)	25 (35.2)	<0.001***
Pyuria (%)	10 (8.6)	13 (18.3)	<0.001***
ANA (positive), n (%)	112 (96.6)	67 (94.4)	0.474
Anti-dsDNA (positive), n (%)	47 (40.5)	42 (59.2)	0.016*
Anti-Sm (positive), n (%)	29 (25.0)	17 (23.9)	0.871

Table 1 continued. General characteristics and clinical manifestations in LN patients with little or no proteinuria according to histologic classification of kidney biopsies.

Variables	Low severity (Classes I and II) (n=116)	High severity (Classes III, IV, and V) (n=71)	p
Anti-RNP (positive), n (%)	52 (44.8)	21 (29.6)	0.038*
Anti-nucleosome (positive), n (%)	32 (27.6)	29 (40.8)	0.077
Patients received PSL prior to admission, n (%)	70 (60.3)	46 (64.8)	0.642
Patients received HCQ prior to admission, n (%)	50 (43.1)	23 (32.4)	0.166
Patients received IA prior to admission, n (%)	46 (39.6)	27 (38.0)	0.878
Cyclophosphamide	34 (29.3)	22 (31.0)	
Mycophenolate	7 (6.0)	3 (4.2)	
Azathioprine	5 (4.3)	2 (2.8)	

Data are shown as mean±standard deviation (SD) or frequencies, as appropriate. * $p<0.05$; ** $p<0.01$; ***: $p<0.001$. SLEDAI – systemic lupus erythematosus disease activity index; PSL – prednisolone; HCQ – hydroxychloroquine; IA – immunosuppressive agents.

160 (85.6%) patients had disease onset between 16–50 years old, 21 (11.2%) patients had pediatric onset (≤16 years), and six (3.2%) patients had late onset (>50 years).

Data collection and measurements

Disease status was determined using the SLE disease activity index (SLEDAI) [29]. Patients with a SLEDAI >10 were considered to have highly active disease. The clinical data included demographics, disease duration, clinical manifestations, immunological measurements, and treatments, all collected from the medical records. Urine and blood samples were routinely obtained at the same time as the renal biopsies. Serum antinuclear antibodies were measured by indirect immunofluorescence on mitotic Hep-2 cells (MBL, Nagoya, Japan). Serum anti-dsDNA antibodies were measured by indirect immunofluorescence assays (EUROIMMUN. AG, Germany). Serum anti-Sm, anti-RNP antibodies, anti-SSA antibodies, anti-SSB antibodies, and antinucleosome were detected using dot blot assays (EUROIMMUN. AG, Germany). C3 and C4 were determined by nephelometry (Bio-red, Beckman Array 360 System, Miami, USA), and hypocomplementemia was defined as C3 <79 mg/dL and/or C4 <16 mg/dL.

Renal marker proteins determined by radioimmunoassay

Levels of serum and urinary β2-MG, UAL, urinary IgG, and urinary α1-MG were measured by radioimmunoassay (Pharmacia RIA kits, Pharmacia Diagnostics, Uppsala, Sweden).

Renal biopsy specimen

Percutaneous renal biopsies were performed under local anesthesia in the left renal pole under ultrasound guidance using automatic 16 gauge needles (Angiotech Surgical Specialties Corp., Wyomissing, PA, USA). Renal biopsy specimens were evaluated by light microscopy (H&E staining, Masson trichrome staining, periodic acid-Schiff staining, and metehamine silver staining), immunofluorescence (IgG, IgA, IgM, C3, and C4 levels), and electron microscopy (ultrastructural changes). Histological renal tissue subtypes were determined according to the 2003 ISN/RPS LN classification [4]. Renal biopsies were analyzed and independently reclassified (Classes I-V) by two renal pathologists and grouped as above (low versus high severity). Activity and chronicity indexes (AI and CI, respectively) were estimated according to previously reported criteria [30]. No serious complication was detected in any patient after renal biopsy.

Statistical analysis

All data were analyzed using SPSS 13.0 (SPSS Inc., Chicago, IL, USA). Continuous variables are expressed as means±standard deviation (SD) and were analyzed using the independent sample t-test. Categorical variables are expressed as proportions and were compared using the chi-square or the Fisher's exact test, as appropriate. Associations between renal marker proteins and histologic classification of kidney biopsies (Classes I and II versus Classes III, IV, and V) were examined by multivariate conditional logistic regression models using the backward stepwise method, including variables that were significantly associated ($p<0.05$) in univariate analyses; UAL and urine IgG were treated as continuous variables, while the others were

Table 2. Renal biopsy findings in 187 Lupus Nephritis (LN) patients with little or no proteinuria.

Class	I	II	III	IV	V	VI
N (%)	5 (2.7)	111 (59.7)	30 (16.1)	32 (17.2)	9 (4.8)	0
Activity index	–	2.57±0.97	4.96±2.08	8.37±3.03	3.73±2.43	–
Chronicity index	–	1.35±1.19	2.75±1.46***	3.18±1.80***	2.54±1.99***	–

In the Class III group, five patients were Class III/V; in the Class IV group, four patients were Class IV/V. *** $p<0.001$ *vs.* Class II.

Table 3. The difference of renal marker proteins in two groups.

Variables	Low severity (Classes I and II) (n=116)	High severity (Classes III, IV, and V) (n=71)	p
Serum β2-MG (µg/ml)	3.49±1.33	3.78±1.49	0.262
Urinary β2-MG (ng/ml)	276±244	303±225	0.517
Urinary albumin (µg/ml)	7.08±10.07	11.60±8.94	0.008**
Urinary IgG (µg/ml)	8.74±8.90	13.21±9.35	0.007**
Urinary α1-MG (µg/ml)	3.03±4.94	4.43±3.75	0.071

Data are shown as mean ±SD. ** $p<0.01$. MG – microglobulin; IgG – immunoglobulin G.

categorized: C3, normal versus low (reference); C4, normal versus low (reference); hypertension without versus with (as reference); anti-dsDNA antibody, negative versus positive (reference); and SLEDAI, 5–9, 10–14, >15 versus 0–4 (reference). Results are reported as odds ratios (OR) with 95% confidence intervals (95% CI). Receiver operating characteristic (ROC) curve analysis, based on the multiple logistic regression model, was used to determine the diagnostic value of the protein markers. Correlations between renal marker proteins and disease activity were analyzed using the Pearson test. Two-sided p values <0.05 were considered significant.

Results

Histological features

The histopathological assessments characteristics of the lesions according to the 2003 ISN/RPS LN classification [4] are presented in Table 2. The most frequent type of histological renal lesion was mesangial proliferative glomerulonephritis (Class II, n=111, 59.7%) followed by diffuse proliferative glomerulonephritis (Class IV, n=32, 17.2%), focal segmental proliferative glomerulonephritis (Class III, n=30, 16.1%), and membranous glomerulonephritis (Class V, n=9, 4.8%) (Table 2). In the Class III group, five patients were Class III/V; in the Class IV group, four patients were Class IV/V. The mean AI was 4.16±2.71 (range 1–14). The mean CI was 2.10±1.66 (range 0–9). The AIs were different among the classes of disease ($p<0.001$). Regarding the CI, significant differences were found between Classes II and III, Classes II and IV, and Classes II and V ($p<0.001$). However,

there was no difference in CI between Classes III, IV, and V ($p>0.05$) (Table 2).

Characteristics of the patients

As shown in Table 1, when comparing the low and high severity groups, there was no difference between the two groups for the female/male ratio, onset age, disease duration, and time between onset of symptoms and LN diagnosis. The time between LN onset and biopsy in the high severity group was longer than in the low severity group (8.0±23.6 versus 20.5±33.2 months, $p=0.022$). Compared with the low severity group, more patients in the high severity group showed fever (62.0% versus 46.6%, $p=0.04$), thrombocytopenia (36.6% versus 17.2%, $p=0.003$), and hypertension (28.2% versus 10.3%, $p=0.002$). No significant difference was observed among the other clinical features. As expected, SLEDAI was higher in the high severity group than in the low severity group (12.5±6.6 versus 7.7±6.5, $p<0.001$). There was no difference in medication prior to admission between the two groups ($p=0.64$ for prednisolone, $p=0.17$ for hydroxychloroquine, and $p=0.88$ for immunosuppressive agents).

Laboratory data are presented in Table 1. All patients had stable serum creatinine levels (<1.5 mg/dL). A higher proportion of patients in the high severity group had low levels of C3 (83.1% versus 50.9%, $p<0.001$), low C4 (71.8% versus 53.4%, $p=0.014$), and positive anti-dsDNA antibodies (59.2% versus 40.5%, $p=0.016$). However, positive anti-RNP antibodies were present in a higher proportion of patients in the low severity group (44.8% versus 29.6%, $p=0.038$). The proportions of

Table 4. Multivariate conditional logistic regression analysis of variables independently associated with high severity renal lesions (class III, IV or V) in LN patients with little or no proteinuria.

Variable	OR	95% CI for OR	p
Urinary albumin	1.417	1.145–1.895	0.001
SLEDAI	2.004	1.264–3.178	0.003

OR – odds ratio; CI – confidence interval; SLEDAI – systemic lupus erythematosus disease activity index.

patients with positive ANA, anti-Sm antibodies, and anti-nucleosome antibodies were not significantly different between the two groups.

Relationship between renal marker proteins and renal LN histopathology

Compared with the low severity group, patients in the high severity group had higher UAL (11.60±8.94 versus 7.08±10.07 µg/mL, *p*=0.008) and urinary IgG (13.21±9.35 versus 8.74±8.90 µg/mL, *p*=0.007) levels (Table 3). There was no difference in serum β2-MG, urinary β2-MG, and urinary α1-MG levels between the two groups.

Multivariate analysis of the renal marker proteins associated with severe renal lesions according to histologic classification of kidney biopsies

For the multivariate analysis, the histologic classification of kidney biopsies (Classes I and II versus Classes III, IV, and V) was set as the dependent variable, and independent variables were low C3, low C4, UAL, urinary IgG, hypertension, anti-dsDNA antibody, and SLEDAI. Results showed that UAL (OR=1.417, 95% CI: 1.145–1.895, *p*=0.001) and SLEDAI (OR=2.004, 95% CI: 1.264–3.178, *p*=0.003) were independently associated with severe renal lesions in patients with LN and little or no proteinuria (Table 4). Therefore, the risk of severe renal damage increased by 1.42-fold for each increase of 1 µg/mL UAL, and increased by 2.00-fold for each categorical increase of SLEDAI.

ROC curve analysis

Figure 1 presents the ROC curve of UAL based on multivariate conditional logistic regression analysis. The AUC was 0.787 (*p*<0.001). The optimal cutoff point of UAL was 7.53 µg/mL, and the sensitivity and specificity were 67% and 82%, respectively, for the detection of high severity (Class III, IV, or V) renal lesions.

Correlations between renal marker proteins and disease activity

Table 5 shows that serum β2-MG, urinary β2-MG, UAL, urinary IgG, and urinary α1-MG were all correlated with SLEDAI
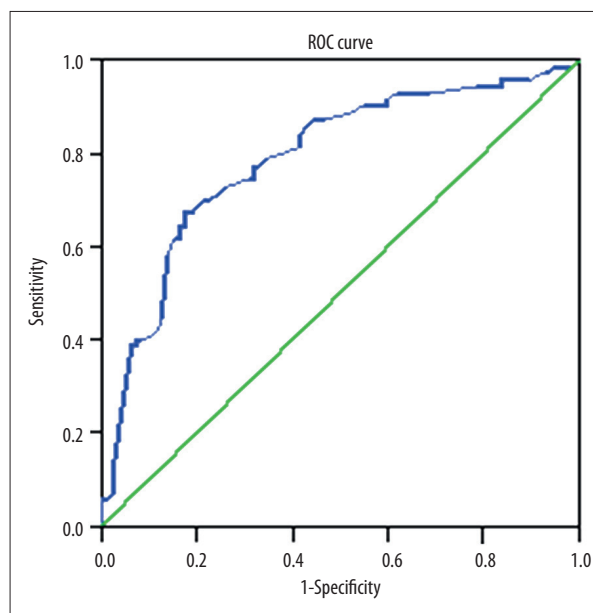

Figure 1. Receiver operating characteristic (ROC) curve showing urinary albumin for the detection of high severity (Classes III, IV, and V) versus low severity (Classes I and II) of renal lesions in lupus nephritis patients with little or no proteinuria. Using a cutoff value of 7.53 µg/mL, urinary albumin showed an area under the ROC curve (AUC) of 0.787, sensitivity of 67%, and specificity of 82%.

(all *p*<0.01), while only UAL (*r*=0.24, *p*=0.01) and urinary IgG (*r*=0.20, *p*=0.03) were correlated with renal biopsy AI. None of these parameters were correlated with renal biopsy CI.

Discussion

The results of this study show that urinary albumin levels and SLEDAI were independently associated with histological severity of renal lesions in patients with LN and little or no proteinuria. These parameters could be used to help select patients for renal biopsy.

Some patients with SLE show evidence of nephritis on renal biopsy despite normal urinary findings and renal function, which is called silent LN (SLN) [15,31]. Although histological renal lesions in patients with SLN are usually mild (i.e.,

Table 5. Correlation between renal marker proteins and disease activity in LN patients with little or no proteinuria.

Variable	SLEDAI		Renal biopsy AI		Renal biopsy CI	
	r	p	r	p	r	p
Serum β2-MG (μg/ml)	0.317	0.005	0.136	0.156	0.032	0.741
Urinary β2-MG (ng/ml)	0.401	<0.001	0.086	0.367	−0.057	0.546
Urinary albumin (μg/ml)	0.374	0.001	0.242	0.010	0.116	0.223
Urinary IgG (μg/ml)	0.396	<0.001	0.204	0.031	0.002	0.985
Urinary α1-MG (μg/ml)	0.349	0.002	0.173	0.069	0.135	0.159

MG – macroglobulin; IgG – immunoglobulin G; SLEDAI – systemic lupus erythematosus disease activity index; AI – activity index; CI – chronicity index.

Class II), diffuse proliferative glomerulonephritis histological changes (Class III or IV), which are related to poor outcomes, could also be present. Zabaleta-Lanz et al. reviewed the literature and reported that 59.8% (122/204) of patients with SLN were ISN/RPS Classes I and II, while 36% were Class III or IV (16% and 20%, respectively) [16]. Wakasugi et al. also reported that 15% of patients with SLE without clinical renal involvement had Classes III and IV LN [32]. In the present study, many patients may have had SLN. On the other hand, prior to admission, some patients had an irregular treatment history of hydroxychloroquine, prednisolone, or immunosuppressive agents that could induce a clinical partial remission to some extent. As shown in Table 1, about 60% of patients had received prednisolone (mostly irregular or discontinuous) and some of them had received immunosuppressive drugs before renal biopsy, which may mask the manifestations of severe nephritis in the early stage.

Disease status was determined using SLEDAI [29] and patients with a SLEDAI >10 were considered to have highly active disease. SLEDAI assigns a weighted score for each of the clinical manifestations of SLE and therefore represents the disease activity of SLE [29,33]. SLEDAI has been shown to be associated with the presence of active LN in some studies [34,35]. In the present study, SLEDAI was independently associated with the severity of kidney lesions in patients with LN and little or no proteinuria. These results are supported by previous studies [36–38].

Proteins of high molecular weight such as IgG (150 kD) are excreted in large quantities when permselectivity of the glomerular capillary wall is more severely disrupted, while proteins of low molecular weight such as α1-MG, β2-MG, or albumin are excreted in large quantities when proximal tubular cells lose their capacity to reabsorb them completely from the tubular lumen [19]. It was found that the urinary excretion of IgG, α1-MG, β2-MG, and albumin may be more sensitive and reliable markers than overall proteinuria for the early detection of renal

involvement in some chronic diseases like diabetes mellitus [19,39–41]. Some studies showed that in patients with glomerular diseases, the urinary excretion of these marker proteins correlated with histologic lesion severity and may more efficiently predict proteinuria quantity, natural disease course, disease outcome, and treatment response [42,43]. Results of the present study suggest that albumin and IgG urinary excretions had a significant relationship with renal histological lesion severity in patients with LN and little or no proteinuria, but only UAL levels were independently associated with renal histological lesion severity in patients with LN and had a good predictive value. These results are supported by previous studies. Indeed, Miranda et al. [44] showed that UAL was associated with the presence of lupus glomerulonephritis (Class II LN). Sui et al. [45] showed that lower serum albumin levels were associated with lesion severity in patients with LN; since serum albumin levels are determined both by liver synthesis and by kidney excretion, this decrease might be due, at least in part, by increased urinary excretion. A deeper investigation of these markers during remission and progression phases, and their relationship to therapeutic response during follow-up, would provide more predictive information.

The correlation analyses revealed that the SLEDAI score was correlated with serum β2-MG, urinary β2-MG, UAL, urinary IgG, and urinary α1-MG. Indeed, the present study, as well as previous studies, revealed that these are markers of kidney damage in a number of pathologies [19,39–43]. As a score representing SLE activity, SLEDAI was associated with these markers.

Of course, many novel promising biomarkers have been evaluated in the studies of LN – for example chemokines, neutrophil gelatinase-associated lipocalin (NGAL), transferrin (TF), and α1-acid-glycoprotein (AGP) [38,46–49]. Currently, however, none has yet been rigorously validated in large-scale longitudinal cohorts of patients with different ethnic backgrounds. Compared with these new biomarkers, the renal marker proteins that were examined in the present study (serum β2-MG,

urinary β2-MG, albumin, IgG, and α1-MG) were easy to measure and have been available and used widely for many years in the clinical setting. However, only UAL was independently associated with severe renal lesions. This lack of association of other factors known to be usually associated with renal pathologies may be due to the sample size or to other factors associated with SLE. Additional studies are necessary to remove the potential effects of confounding factors. The univariate analyses also suggest that some clinical features like hypertension, low C3 and C4 levels, positive anti-dsDNA antibodies, and high disease activity were associated with significant renal lesions in this population of patients with LN, but since these parameters were not identified by the multivariate analysis, their usefulness remains to be determined. Results suggest that for patients with little or no proteinuria, having abnormal predictive indicators, clinical features, and urinary protein markers indicates renal involvement. Of course, these biomarkers will not substitute for renal biopsy. It is considered unethical to perform a renal biopsy in patients without overt symptoms of LN, but an significant proportion of patients have renal damage despite the lack of symptoms. Identifying these patients before symptom onset could provide an opportunity to treat them before disease progression to higher classes. Biopsy will still be necessary to confirm the diagnosis. Therefore, determining early biomarkers associated with LN, even in the absence of signs or symptoms, is of clinical importance.

The present study has some limitations. The sample size was small and from a single center. The cross-sectional nature of the study prevented the analysis of the associations between biomarkers and response to therapy. There is a possibility of selection bias, since these patients were all seen at the Rheumatism Department because they mostly had normal renal function, while patients hospitalized for renal failure are usually treated at the Nephrology Department. In addition, one of the main symptoms was fever, which is supported by other Chinese studies [50,51], but it was impossible to differentiate SLE-related fever from infection fever. Data for cellular components in urine samples were not obtained by manual microscopy, but using an automated analysis of urinary sediment by flow cytometry, which may introduce some bias. Other markers such as the urinary albumin/creatinine ratio could be explored, but this parameter was not measured during the study period (2005–2010) and it requires repeated measurements, which is impractical in the economic context of China. Finally, only a limited panel of biomarkers was assessed, and additional markers could be explored, such as tumor necrosis factor α (TNF-α), TNF-α soluble receptor p75, and interleukin (IL)-6 [52]. Of course, many novel promising biomarkers have been evaluated in the studies of LN, such as neutrophil gelatinase-associated lipocalin (NGAL), transferrin (TF), and α1-acid-glycoprotein (AGP) [38,46–49]. Additional studies are necessary to explore these markers and to determine the best ones in predicting kidney damage in LN with low or no proteinuria. Large multicenter prospective studies are needed to investigate LN and its correlation to urine biomarkers and clinical markers like serum complements and auto-antibodies.

Conclusions

UAL and SLEDAI are independently associated with renal histological lesion severity. These parameters could be used to help identify patients needing a renal biopsy.

Conflicts of interest

The authors declare that they have no conflicts of interest.

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