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# Elevation of Erythrocyte Sedimentation Rate and C-reactive Protein Reflects Renal Interstitial Inflammation in Drug-Induced Acute Tubulointerstitial Nephritis

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# Abstract

Background: A renal biopsy is needed to define active inflammatory infiltrate and guide therapeutic management in drug-induced acute tubulointerstitial nephritis (D-ATIN). However, factors like various contraindications, refused informed consent or limited technical support, would stop the process. It is thus of great importance to explore some approaches that could deduce the probable pathologic changes.

Methods: 64 biopsy-proven D-ATIN patients were enrolled from a prospective cohort of ATIN in Peking University First Hosptial. Systemic inflammation score (SIS) was developed based on the levels of CRP and ESR at biopsy and patients were divided into high-SIS, median-SIS, and low-SIS groups. The demographic data, clinicopathologic features, and renal outcome were compared.

Results: SIS correlated positively with inflammatory cell infiltration and inversely with interstitial fibrosis. The number of each kind of interstitial inflammatory cells increased significantly with the increase in SIS. The proportions of neutrophils and plasma cells were the highest in patients with high-SIS than the other two groups. Prednisone (30–40 mg/day) was prescribed in all the patients. High-SIS group tended to have more favorable renal restoration than the other two groups. By 12 months post-renal biopsy, decreased eGFR (<60 mL/min/1.73 m 2 ) was observed in 70.0% of medium-SIS patients, 33.3% of high-SIS patients, and 29.4% of low-SIS patients.

Conclusion: SIS was positively correlated with active tubulointerstitial inflammation and therefore could help to aid therapeutic decisions in D-ATIN.


# 1. Background

Acute tubulointerstitial nephritis (ATIN) is a common renal lesion histopathologically characterized by inflammation and edema of the renal interstitium. It is responsible for 15–27% of acute kidney injury (AKI)[1–5], with drugs being the most common causes [6–10]. Unlike ischemic or toxic AKI which usually induces acute tubular injury and presents an abrupt decline in renal function, patients with D-ATIN sometimes have insidious renal dysfunction and are therefore more likely to be delayed recognized. A renal biopsy is needed to make a definitive diagnosis of ATIN and reveals the activity and severity of interstitial inflammation that usually directs immunosuppressive treatment[11, 12 13]. However, factors like various contraindications, refused informed consent or limited technical support, would stop the process of renal biopsy. It is thus of great importance, for those patients clinically suspected ATIN whereas renal biopsy cannot be conducted, to explore some approaches that could deduce the probable pathologic changes and indicate the severity of interstitial inflammatory cell infiltration, which could therefore help make therapeutic decisions.

Erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) are traditional inflammatory markers that have been used to help assess the activity of inflammation in various diseases such as systemic lupus erythematosus[14–17], rheumatoid arthritis[18, 19], and vasculitis[20]. However, there is still a lack of

knowledge about the relevance of these systemic inflammatory parameters to renal inflammation. The current study was performed in a prospective cohort of drug-induced ATIN (D-ATIN) patients. The clinical-pathological features and renal recovery of patients with different levels of CRP and ESR were compared. Associations between systemic inflammation and renal tubulointerstitial inflammation as well as the long-term renal outcome were further analyzed.

# 2. Methods

## 2.1 Patients

The study was approved by the Committee on Research Ethics of Peking University First Hospital. Patients who were clinicopathologically diagnosed as D-ATIN from January 1$^{st}$, 2005 to December 31$^{st}$, 2013 and followed up for at least 12 months were screened in a prospective cohort of ATIN in Peking University First Hospital as previously described[21]. The diagnosis of D-ATIN was made based on previously described criteria[22]. A presence of prominent interstitial inflammation in the nonfibrotic cortex and tubulitis was essential for the pathologic diagnosis of ATIN. Those who were younger than 14 years, with renal glomerular diseases, malignancy, systemic autoimmune diseases, or concurrent infections, were excluded. Altogether, 64 D-ATIN patients were enrolled in the current study.

## 2.2 Clinical parameters evaluation and grouping for systemic inflammation

Clinical parameters and laboratory data were documented. Acute kidney disease (AKD) were defined using the Kidney Disease: Improving Global Outcomes (KDIGO) criteria[23] and consensus report of the Acute Disease Quality Initiative (ADQI) 16 Workgroup[24]. ESR and CRP were categorized by quartile and ranked 0~3 points respectively (Table 1). Systemic inflammatory score (SIS) was calculated by combining the points of ESR and CRP, and patients were classified into 3 systemic inflammation groups, including low-SIS (score 0–1), medium-SIS (score 2–3) and high-SIS (score 4–6) groups.

## 2.3 Evaluation of renal pathologic features

All kidney biopsy tissues were processed for light microscopy, immunofluorescence, and electron microscopy. Semiquantitative scores for interstitial edema, infiltration, fibrosis, tubulitis and tubular atrophy were assessed by two pathologists referring to a modification of the Banff Working Classification[25, 26]. Scores 1–3 are considered to correspond to mild, moderate, and severe, respectively. The activity index was the total of the scores for interstitial edema, infiltration and tubulitis. The chronicity index was the total of the scores for interstitial fibrosis and tubular atrophy.

## 2.4 Immunofluorescence staining of infiltrated inflammatory cells

Infiltrating cells were identified by immunofluorescence staining with antibodies against CD3 (T lymphocytes, 1:100, ZM–0417, ZSGB-BIO), CD20 (B lymphocytes, 1:100, ZA–0293, ZSGB-BIO), CD38 (plasma cells, 1:250, ab108403, Abcam), CD68 (monocytes/macrophages, 1:500, ZM–0060, ZSGB-BIO), and neutrophil elastic protease antibodies (neutrophils,1:200, GWB–8F72C4, Genway-Bio), and counted at 400x magnification. Eosinophils were detected with hematoxylin and eosin staining and counted at 200x magnification. The quantitation of the tubulointerstitial infiltration was determined by averaging the counts of five randomly selected fields. The mean values are expressed as cells per millimeter squared.

## 2.5 Follow-up, renal recovery and renal outcome

Serum creatine (SCr) was routinely performed during the follow-up periods. Renal recovery was based on SCr at 6 months post-biopsy. Complete recovery was defined as improvement in SCr level to within 25% of its baseline (or to 133mol/L if the baseline was not available); partial recovery as a >50% decrease in SCr level from its peak value but not reaching within 25% of its baseline value; and no recovery as the failure to meet the criteria for complete or partial recovery or remaining on renal replacement treatment (RRT). Renal outcomes were defined by the estimated glomerular filtration rate (eGFR) at 12 months post-biopsy. eGFR was calculated by the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) equation[27] and expressed as milliliters per minute per 1.73 $m^2$.

## 2.6 Statistical Analysis

Analyses were performed using SPSS 22.0 Statistics software (IBM Corp., Armonk, NY) and GraphPad Prism version 6 (GraphPad Software, San Diego, CA). Categorical variables were expressed as counts and percentages. Continuous data accorded with normal distribution were presented as mean and s.d. and those with abnormal distribution were presented as median and interquartile range. To assess group differences, one-way analyses of variance and Chi-squared analyses were conducted. Bonferroni post hoc comparisons were computed when significant differences emerged. Spearman's rank correlation coefficient was used as a measure of correlations between SIS and clinicopathological parameters. Multiple linear regression analysis was computed with the activity index as the dependent variable, and the following variables as explanatory variables: SIS, sex, age, disease course, hemoglobin and eGFR levels at biopsy. A two-sided $P<0.05$ was considered statistically significant.

# 3. Result

## 3.1 Baseline demographic and clinical data

As expressed in Table 2, the average age of 64 D-ATIN patients was 45.7±12.7 years, with a female predominance (41/64, 64.1%). The interval from initiation of drug use to the diagnostic biopsy was 50 (30–83) days. The majority of the cases (62/64, 96.9%) were identified as AKD, 2 cases (3.1%) were classified as CKD. 16 cases (25%) required and initiated RRT before the biopsy. 6 cases (20.7%) were oliguric. 15 cases (23.4%) had allergic history. Common clinical features included digestive symptoms (62.5%), weakness (53.1%), fever (53.1%) and rash (14.1%).

The number of medications that were related to D-ATIN was identified as one kind in 39 patients (60.9%), two kinds in 20 patients (31.3%), and three or more kinds in 5 cases (7.8%). The most common culprit drugs were antibiotics (39/64, 60.9%), followed by herbal medicine (23/64, 35.9%), nonsteroidal anti-inflammatory drugs (NSAIDs) (22/64, 34.4%), proton pump inhibitors (2/64, 3.1%), antipsychotic (1/64, 1.6%), and antiviral agent (1/64, 1.6%)etc.

# 3.2 Clinical relevance of SIS in D-ATIN patients

Of the total 64 D-ATIN patients, ESR was elevated in 55 cases (85.9%) with an average level of 63.8 (37.5–88.0) mm/hr. CRP was elevated in 35 cases (54.7%) with an average value of 10.0 (4.4–24.4) mg/L. SIS evaluated by both ESR and CRP levels positively correlated with SCr values at renal biopsy (r = 0.376，*P* = 0.002), leukocyturia (r = 0.344，*P* = 0.001) and C3 levels(r = 0.547，*P*<0.001) (Supplementary Table 1).

Based on SIS values, 17 patients were grouped as low-SIS, 20 as medium-SIS and 27 as high-SIS. There was no significant difference in age, gender, allergic manifestations, or causal medications among the three groups of patients. Patients in the low-SIS group had mildest kidney injury with lowest SCr levels at renal biopsy (median value: 154μmol/L, *P* = 0.002), lowest RRT rate (5.9%, *P* = 0.021) and highest hemoglobin concentrations (113.1±11.8g/L, *P* = 0.001). It is interesting to note that patients in the medium-SIS group tended to have higher peak SCr levels (median 417 vs 324μmol/L) and RRT rates (45.0% vs 22.1%) than those with high-SIS, yet their disease courses were relatively longer (median 60 vs 51 Days) with lower levels of SCr at biopsy (median 275 vs 306μmol/L). In addition, patients in the high-SIS group had significantly higher C3 levels (1.3±0.2 vs 1.0±0.2 mg/L, *P*<0.001) with a greater prevalence of leukocyturia (82% vs 55%, P = 0.003) compared to those with medium-SIS(Table 2).

# 3.3 Pathological relevance of SIS in D-ATIN patients

Compared to the patients with low-SIS and medium-SIS, those in the high-SIS group had the highest degree of interstitial inflammation (*P*<0.001) and the lowest degree of interstitial fibrosis (*P* = 0.038) (Table 3). SIS correlated positively with renal interstitial inflammatory cell infiltration (r = 0.64; *P*<0.001) and interstitial edema (r = 0.25; *P* = 0.046), and inversely with interstitial fibrosis (r = −0.33; *P* = 0.009) (Supplementary Table 1). Multiple linear regression analysis demonstrated that only SIS had a significant correlation with renal activity index(β coefficient = 0.383, *P*<0.001).

We next investigated renal interstitial inflammatory cell types through immunofluorescence staining. The number of each kind of interstitial inflammatory cells increased significantly with the increase in SIS (Table 3 and Figure 1). When focusing on the constitution of inflammatory cells, the proportions of neutrophils (7.9% vs 2.6% in medium-SIS vs 1.2% in low-SIS; $P$<0.001) and plasma cells (12.9% vs 9.0% in medium-SIS vs 9.4% in low-SIS; $P$ = 0.023) were the highest in patients with high-SIS than the other two groups. There was no significant difference in the proportions of T lymphocytes, B lymphocytes or macrophages among the three groups of patients. Eosinophils, which favor a diagnosis of drug-induced ATIN, were also of the greatest number in the high-SIS group (median value: 3.9 vs 0.8 in medium-SIS vs 0.4 in low-SIS; $P$<0.001).

## 3.4 Treatment and outcome among three groups with different SIS

As shown in Table 4, prednisone was prescribed at a dosage of 30–40 mg/day in all the patients. Additional immunosuppressive agents, such as mycophenolate, azathioprine and cyclophosphamide, were used in 28.1% (18/64) of patients with no significant difference among the three groups ($P$ = 0.304). Methylprednisolone impulse was performed in 23.4% (15/64) of overall patients, and the proportion was similar in patients in both groups of medium-SIS and high-SIS (approximately 30.0%). None of the low-SIS patients received methylprednisolone impulse therapy.

Patients were followed up for at least 12 months (12–132 months, median 43 months). High-SIS group tended to have more favorable renal restoration than the other two groups(Figure 2). At 6 months post-biopsy, complete recovery was achieved in 70.4% of high-SIS patients, 55.0% of medium-SIS patients, and 64.7% of low-SIS patients ($P$ = 0.407). Decreased eGFR (<60 mL/min/1.73 $m^2$) was observed in 33.3% of high-SIS patients, 70.0% of medium-SIS patients, and 29.4% of low-SIS patients ($P$ = 0.016) at 12 months post-biopsy, (Table 4).

30 patients with severe renal dysfunction at the time of biopsy (eGFR <20 mL/min/1.73 $m^2$) were divided into high-score and low-score subgroups based on SIS. The eGFR values at biopsy were similar in the two subgroups(12.1±5.6 in low-score vs 10.6±5.1 mL/min/1.73 $m^2$ in high-score, $P$ = 0.442). At 12 months post-biopsy, the eGFR values were higher in high-score subgroup ( 59.5±16.5 vs 49.2±13.7 mL/min/1.73 $m^2$ in low-score, $P$ = 0.074), and the proportion of eGFR less than 60 mL/min/1.73 $m^2$ was significantly smaller in high-score subgroup(46.7% vs 86.7% in low-score, $P$ = 0.025).

# 4. Discussion

D-ATIN is a relatively common cause of AKI. Previously, ESR and CRP had been reported to be significantly elevated in D-ATIN patients[13, 28]. The current study first demonstrated that SIS evaluated by ESR and CRP was correlated with active renal tubulointerstitial inflammation and renal restoration in a

prospective cohort of D-ATIN, and therefore could help to aid therapeutic decisions when a renal biopsy is not acceptable or cannot be performed serially in this disease condition.

Patients with D-ATIN often present with a relatively insidious onset and a disease process of subacute renal dysfunction, with oliguria not commonly seen[13]. Therefore, a delayed diagnosis was likely to be encountered especially when patients are initially attended at non-nephrology departments, as is showed in our study where the median time course from the initial use of suspicious drugs to diagnostic renal biopsy was 50 days with some cases even exceeded 3 months. Once acute interstitial inflammation sets in, it can progress rapidly to a less reversible, more destructive fibrogenic process[29]. Therefore, the delay in diagnosis and treatment would result in complexity in the pathophysiologic process containing both active inflammation and fibrotic lesions in patients with D-ATIN. It is crucial to make a treatment decision on evidence that reflects renal interstitial active inflammation, yet renal biopsy, the golden standard for histological evaluation, is invasive and not acceptable for all patients. Following multivariate analysis, we observed that systemic inflammatory markers closely correlated with renal inflammation regardless of disease course. More importantly, it has been reported that D-ATIN patients might experience recurrent kidney injury during long-term follow-up due to various medications[21]. Elevated systemic inflammatory markers combined with abnormal urinary markers during follow-up may play an important role in reflecting the degree of renal inflammation and providing the proper management among those patients.

During follow-up, for patients with similar eGFR values at biopsy, better renal outcomes were achieved in those with higher systemic inflammatory scores, suggesting that SIS might serve as an indicator of renal outcome. As for pathological findings, SIS correlated positively with renal interstitial inflammatory cell infiltration especially with neutrophils and eosinophils that participate in the acute phase of inflammation[30], and correlated inversely with interstitial fibrosis, which suggests its ability in reflecting the activity of renal interstitial inflammatory injury in D-ATIN. A positive correlation was also observed between SIS and plasma cell ratio. Plasma cell infiltration was found to be positively correlated with CRP and tubulointerstitial inflammation scores in antineutrophil cytoplasmic autoantibody-associated vasculitis[31]. The radical role of plasma cells in D-ATIN remains unclear, but we suppose that aggregated plasma cells in the kidney may play a role in local inflammation in the early phase. In summary, our findings indicate that SIS could serve as a non-invasive biomarker for an ongoing inflammatory process and active kidney injury in D-ATIN which might benefit from prompt treatment.

Our study has limitations related to the retrospective observational design. Selection bias originates from a single-center study and the inclusion of patients with biopsy-proven D-ATIN, which would miss less severe cases. CRP and ESR are non-specific markers that could be elevated in various conditions such as infection, systemic autoimmune diseases, etc. Therefore the introduction of SIS in D-ATIN should be implemented after excluding these conditions.

# 5. Conclusions

Our study firstly demonstrated the relationship between systemic inflammation and local renal inflammation in D-ATIN. SIS based on ESR and CRP could serve as a marker of the activity of tubulointerstitial inflammation and assist decision-making in immunosuppressive therapy of D-ATIN.

## List Of Abbreviations

D-ATIN: drug-induced acute tubulointerstitial nephritis

CRP: C reactive protein

ESR: erythrocyte sedimentation rate

SIS: systemic inflammation score

HPF:high power field

SCr:serum creatine

AKD: acute kidney disease

KDIGO: Kidney Disease: Improving Global Outcomes

ADQI: the Acute Disease Quality Initiative

RRT: renal replacement treatment

eGFR: estimated glomerular filtration rate

CKD-EPI: Chronic Kidney Disease Epidemiology Collaboration

CKD: chronic kidney disease

## Declarations

# Ethics approval and consent to participate

The study was performed in compliance with the Declaration of Helsinki and approved by the Ethics Committee of Peking University First Hospital(approval number 2017[1280]). Written informed consent for obtaining tissue, blood and urine samples were obtained from each participant or their parents or guardian (for participants under 16 years old).

# Consent for publication

The results presented in this paper have not been published previously in whole or part, except in abstract form at 3rd Asia Pacific AKI CRRT 2019 Congress. We have obtained the necessary permission from the copyright holder.

# Availability of data and materials

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

# Competing interests

The authors declare that they have no competing interests.

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# Authors’ contributions

XZZ collected the data, interpreted the statistical results, and wrote the first draft. YHG and DMX collected the data. TS, XJZ, GL and LY contributed to the design of the protocol and revised the manuscript. SXW, JWH, PPS and YJ performed the histological examination of the kidney. All authors read and approved the final manuscript.

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# Tables

**Table 1. Scoring system based on values of ESR and CRP**

| CRP (mg/dL) | ESR (mm/hr) | | | |
|---|---|---|---|---|
| | <38 (0 Point) | 38-62 (1 Points) | 62-88 (2 Points) | ≥88 (3 Points) |
| <4.4(0 Point) | 0 | 1 | 2 | 3 |
| 4.4-10.0 (1 Points) | 1 | 2 | 3 | 4 |
| 10.0-24.4 (2 Points) | 2 | 3 | 4 | 5 |
| ≥24.4 (3 Points) | 3 | 4 | 5 | 6 |

**Table 2. Demographic and clinical features in different SIS groups**

| Variables | Total N=64 | Low-SIS N=17 | Medium-SIS N=20 | High-SIS N=27 | *P*-value |
|---|---|---|---|---|---|
| Female, n(%) | 41 (64.1) | 8(47.1) | 15(75.0) | 18 (66.7) | 0.197 |
| Age (year) | 45.7±12.7 | 44.1±16.5 | 46.7±11.1 | 45.8±11.6 | 0.827 |
| Disease course (day)[a] | 50 (30, 83) | 50 (33, 90) | 60 (33, 90) | 51 (30, 60) | 0.444 |
| Thyroid disease, n(%) | 9 (14.1) | 4(23.5) | 4(20.0) | 1 (3.7) | 0.088 |
| Allergic history, n(%) | 15 (23.4) | 7 (41.2) | 4(20.0) | 4 (14.8) | 0.136 |
| Fever, n(%) | 34 (53.1) | 8 (47.1) | 9(45.0) | 17 (63.0) | 0.400 |
| Rash, n(%) | 9 (14.1) | 3 (17.6) | 2 (10.0) | 4 (14.8) | 0.786 |
| AKD, n(%) | 62 (96.9) | 15(88.3) | 20(100.0) | 27 (100.0) | 0.058 |
| RRT, n(%) | 16 (14.1) | 1 (5.9) | 9(45.0)* | 6 (22.1) | 0.021 |
| Oliguria, n(%) | 6 (14.1) | 0 (0) | 2 (10.0) | 4 (14.8) | 0.124 |
| Suspected drug, n(%) | | | | | |
| Antibiotics | 39 (60.9) | 11 (64.7) | 8(40.0) | 20 (74.1) | 0.057 |
| NSAIDs | 22 (34.4) | 7 (41.2) | 5(25.0) | 10 (37.0) | 0.545 |
| Herbal medicine | 23 (35.9) | 7 (41.2) | 9(45.0) | 7 (25.9) | 0.351 |
| Multiple drugs | 25 (39.1) | 9 (52.9) | 5(25.0) | 11 (40.7) | 0.260 |
| Laboratory tests | | | | | |
| SCr at peak (μmol/L) | 321 (221, 534) | 285 (203, 437) | 417(226, 647) | 324(220, 523) | 0.204 |
| SCr at biopsy (μmol/L) | 249 (163, 418) | 154(130, 246) | 275(205, 585)* | 306 (182, 480)* | 0.002 |
| Hematuria, n (%) | 17 (26.6) | 3 (17.6) | 7(35.0) | 7 (25.9) | 0.490 |
| Leukocyturia, n (%) | 39 (60.9) | 6 (35.3) | 11 (55.0) | 22 (81.5)* | 0.008 |
| UTP (g/24h) | 1.1 (0.7, 1.7) | 0.7 (0.4, 1.5) | 1.2 (0.7, 1.8) | 1.2 (1.0, 1.7) | 0.129 |
| U-NAG (U/L) | 23 (14, 48) | 31 (16, 67) | 19 (12, 31) | 23 (16, 52) | 0.289 |
| U-a1MG (mg/L) | 193 (101, 48) | 160 (12, 232) | 160 (59, 314) | 200 (153, 232) | 0.305 |
| U-mAlb (mg/L) | 70 (38, 146) | 46 (15, 125) | 62(40, 130) | 78 (60, 184) | 0.128 |

| | | | | | |
|---|---|---|---|---|---|
| Renal glycosuria, n (%) | 51 (79.7) | 12 (70.6) | 16 (80.0) | 23 (85.2) | 0.513 |
| U-Osm decrease, n(%) | 45 (70.3) | 9 (52.9) | 16 (80.0) | 20 (74.1) | 0.170 |
| RTA, n (%) | 37 (57.8) | 4 (23.5) | 15 (75.0) * | 18 (66.7) * | 0.003 |
| Hemoglobin (g/L) | 102.0±15.8 | 113.1±11.8 | 98.4±18.0* | 97.7±13.3* | 0.001 |
| Hypokalemia, n (%) | 30 (46.9) | 7 (41.2) | 10 (50.0) | 12 (44.4) | 0.853 |
| IgG (g/L) | 15.6±4.4 | 15.0±3.9 | 15.3±3.3 | 16.1±5.4 | 0.681 |
| C3 (mg/L) | 1.1±0.3 | 0.9±0.2 | 1.0±0.2 | 1.3±0.2*# | <0.001 |

Abbreviations: SIS, systemic inflammatory score; NSAIDs, non-steroidal anti-inflammatory drugs; SCr, serum creatinine; eGFR, estimated glomerular filtration rates; UTP, urinary total protein; U-KIM1, urinary kidney injury molecular 1; U-NAG, urinary N-acetyl-β-D-glucosaminidase; U-α1MG, urinary α1 microglobulin; U-mAlb, urinary microalbumin; U-Osm, urinary osmolality; RTA, renal tubular acidosis; IgG, immunoglobulin G; C3, complement 3.

[a] Disease course was defined as the interval from initiation of drug use to the diagnostic biopsy；[b] Other drugs included proton pump inhibitors, antipsychotic, antiviral agent, and neuromuscular release agent.

[*] compared with low-SIS group, $P<0.05$；[#] compared with medium-SIS group, $P<0.05$

Normal range：U-NAG (0.3-12) U/L；U-α1MG (0-12)mg/L；U-mAlb (0-19) mg/L；IgG (7.2-16.9) g/L；C3 (0.6-1.5)mg/L

**Table 3. Pathology features in different SIS groups**

| Variables | Total N=64 | Low-SIS N=17 | Medium-SIS N=20 | High-SIS N=27 | *P*-value |
|---|---|---|---|---|---|
| ***Semiquantitative pathologic score*** | | | | | |
| Activity index | 4 (3, 5) | 3 (2, 3) | 4 (3, 4) | 4 (4, 5)*# | <0.001 |
| Interstitial edema | 1 (0.25, 1) | 1 (0, 1) | 1 (0, 1) | 1 (1, 1) | 0.262 |
| Interstitial inflamation | 3 (2, 4) | 2 (2, 3) | 3 (2, 4) | 4 (3, 4) *# | <0.001 |
| Tubulitis | 0 (0, 0) | 0 (0, 0) | 0 (0, 0) | 0 (0, 0) | 0.479 |
| Chronicity index | 0 (0, 2) | 0 (0, 1) | 0 (0, 2) | 0 (0, 1) | 0.733 |
| Interstitial fibrosis | 0 (0, 0) | 0 (0, 1) | 0 (0, 2) | 0 (0, 0) # | 0.038 |
| Tubular atrophy | 0 (0, 1) | 0 (0, 1) | 0 (0, 1) | 0 (0, 1) | 0.448 |
| ***Interstitial inflammatory cell counts*** | | | | | |
| Total cells [a] | 391.5±135.5 | 307.0±147.8 | 373.2±94.7 | 452.8±125.8*# | 0.002 |
| T lymphocytes | 173.9±63.6 | 137.6±72.3 | 173.2±62.9 | 194.7±50.8* | 0.024 |
| B lymphocytes | 45.4(30.4, 61.9) | 34.7 (14.4, 41.6) | 47.3 (30.8, 58.8) | 57.3 (36.9, 70.2)* | 0.020 |
| Monocytes/macrophages | 101.8±34.8 | 77.6±35.9 | 90.2±29.6 | 123.2±24.1*# | <0.001 |
| Plasma cells | 43.8±24.3 | 30.3±20.7 | 31.8±19.0 | 59.4±20.6*# | <0.001 |
| Neutrophils | 15.4 (4.9, 32.9) | 2.9 (1.7, 13.8) | 10.0 (4.8, 19.2) | 33.4 (18.9, 49.9)*# | <0.001 |
| Eosinophils [b] | 2.0 (0.5, 5.1) | 0.4 (0.1, 1.7) | 0.8 (0.1, 2.3) | 3.9 (2.2, 9.4) *# | <0.001 |
| ***Percentages of Interstitial inflammatory cells (%)*** | | | | | |
| T lymphocytes | 44.3±9.0 | 46.9±8.5 | 46.7±10.1 | 41.1±7.8 | 0.064 |
| B lymphocytes | 11.7 (9.0, 14.3) | 10.6 (8.8, 14.5) | 12.9 (9.5, 14.8) | 11.2 (86, 13.8) | 0.475 |
| Monocytes/macrophages | 25.9 (21.3, 29.9) | 25.8 (21.5, 30.9) | 25.7 (19.9, 30.1) | 26.4 (23.9, 29.2) | 0.800 |
| Plasma cells | 10.9±5.2 | 9.4±4.4 | 9.0±5.6 | 12.9±4.7*# | 0.023 |

| | | | | | |
|---|---|---|---|---|---|
| Neutrophils | 4.3 (1.6, 7.8) | 1.2 (0.9, 5.4) | 2.6 (1.7, 4.7) | 7.9 (4.7, 11.7)*# | <0.001 |

Abbreviations: SIS, systemic inflammatory score;

[a] Total cells count was the sum of T lymphocytes、B lymphocytes、macrophages、plasma cells and neutrophil under 400´ magnification.

[b] Eosinophils were counted under 200´ magnification.

*: compared with low-SIS group, *P*<0.05

#: compared with medium-SIS group, *P*<0.05

**Table 4. Treatment and renal outcome in different SIS groups**

| Variables | Total N=64 | Low-SIS N=17 | Medium-SIS N=20 | High-SIS N=27 | *P*-value |
|---|---|---|---|---|---|
| Immunosuppressive treatment, n (%) | | | | | |
| Prednisone only | 41 (64.1) | 14(82.4) | 11(55.0) | 16 (59.3) | 0.178 |
| Methylprednisolone impulse | 14 (21.9) | 0 (0) | 6(30.0)* | 8(29.6)* | 0.007 |
| Immunosuppressive medications | 18 (28.1) | 3 (17.6) | 8(40.0) | 7 (25.9) | 0.304 |
| Renal recovery at 6 months post-biopsy、n、%、 | | | | | |
| Complete | 41 (64.1) | 11 (64.7) | 11 (55.0) | 19 (70.4) | 0.407 |
| Partial | 22 (34.4) | 5 (29.4) | 9(45.0) | 8 (29.6) | |
| None | 1 (1.6) | 1(5.9) | 0(0) | 0(0) | |
| Renal outcome evaluated by eGFR (mL/min/1.73 $m^2$) | | | | | |
| ≥60 | 36 (56.3) | 12 (70.6) | 6(30.0)* | 18(66.7)# | 0.016 |
| <60 | 28 (43.8) | 5 (29.4) | 14(70.0) | 9(33.3) | |

Abbreviations: SIS, systemic inflammatory score; eGFR, estimated glomerular filtration rates; CKD, chronic kidney disease; CRP, C reactive protein; ESR, erythrocyte sedimentation rate;

$\Delta eGFR_{1year\text{-}biopsy} = eGFR_{6month\ after\ biopsy} - eGFR_{biopsy}$.

Rate of $\Delta eGFR_{6months\text{-}biopsy} = \Delta eGFR_{6months\ -biopsy} / eGFR_{biopsy}$

*: compared with low-SIS group, P<0.05

#: compared with medium-SIS group, P<0.05

# Figures

**Figure 1**

Histological findings of drug-induced acute tubulointerstitial nephritis (A)Severe interstitial inflammatory infiltrates with preserved glomerulus (Periodic methenamine silver and Masson Trichrome staining, ×200). (B) Interstitial infiltration by mononuclear cells，eosinophils(white arrow) and neutrophils(black arrow) (hematoxylin-eosin staining, ×400). (C) Double immunofluorescence staining for T lymphocytes(green) and B lymphocytes(red)(×400). (D-F) Immunofluorescence staining for neutrophils (red) , plasma cells (green) and eosinophils (red) (×400).

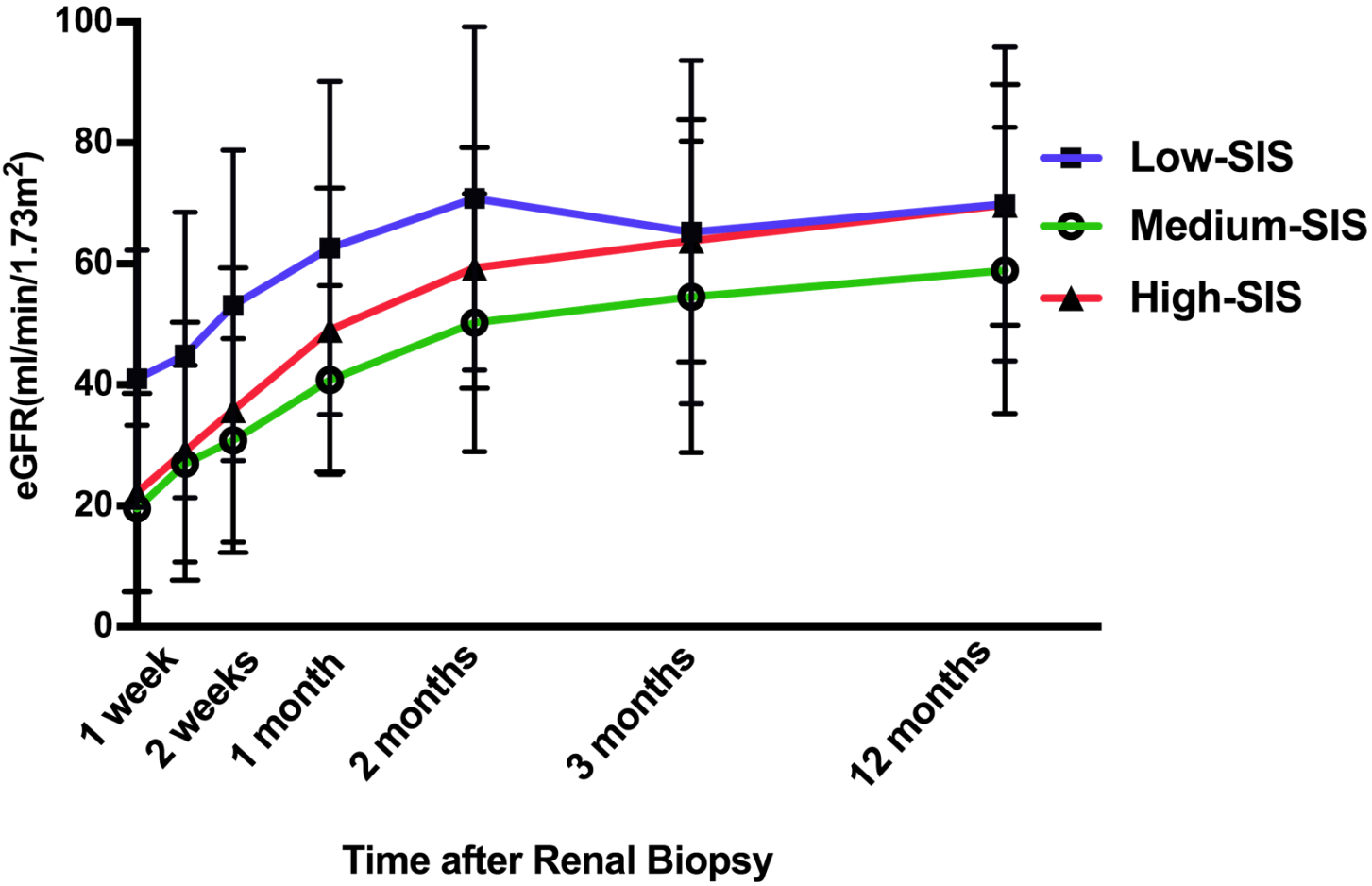


**Figure 2**

The restoration of renal function in patients with low-SIS (n=17), medium-SIS (n =20) and high-SIS (n=27) during the first year post-renal biopsy. Patients in both groups of medium-SIS and high-SIS had

significantly renal dysfunction at the time of renal biopsy, and high-SIS patients presented more rapidly renal function restoration. Low-SIS patients had modest renal dysfunction and modest restoration of renal function.

## Supplementary Files

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- SupplementaryTable.docx