Original Paper

Inflammatory Mediators in Kidney Disease: A Focus on IL-6 in SLE-Associated CKD

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Introduction. Systemic lupus erythematosus (SLE) is an autoimmune condition that can lead to severe renal impairment, including chronic kidney disease (CKD) or end-stage kidney disease (ESKD). This study aimed to evaluate the serum levels of key pro-inflammatory cytokines which are well-known to mediate tissue injury and fibrosis in the context of kidney diseases—interleukin-1 beta (IL-1β), IL-6, tumor necrosis factor-alpha (TNF-α), and IL-12—in CKD and ESKD patients with and without SLE, as well as in healthy controls, to explore their role in disease progression.

Methods. This cross-sectional study was conducted over a 15-month period in three governmental medical centers of Golestan Province, Iran (Sayyad Shirazi Hospital, 5th Azar hospital, and Al-Jalil hospital). A total of 118 individuals (age 18-65 years) were enrolled and divided into three groups:38 CKD or ESKD patients with SLE, 40 CKD or ESKD patients without SLE, and 40 healthy controls. For subgroup analyses, CKD (stages3-4) and ESKD (stage 5) patients were considered separately. Those patients meeting the American College of Rheumatology criteria for SLE, while excluding those with infections, malignancies, or other autoimmune disorders were selected. Blood samples were collected, and serum cytokine levels were measured using enzyme-linked immunosorbent assay (ELISA). Statistical analysis was performed using the Kruskal-Wallis and Mann-Whitney U tests, with $P \leq .05$ considered significant.

Results. CKD patients with SLE showed significantly higher serum levels of IL-1β, TNF-α, and IL-12 compared to CKD patients without SLE and healthy controls ($P < .05$). IL-6 levels were notably higher in CKD patients compared to ESKD patients, irrespective of SLE status ($P < .05$). ESKD patients exhibited increased IL-1β, TNF-α, and IL-12 levels, while IL-6 levels were lower than those in CKD patients.

Conclusion. IL-6 appears to play a crucial role in the pathogenesis of kidney disease, particularly in patients with SLE, and may serve as a therapeutic target. The differential regulation of cytokines in CKD and ESKD suggests that inflammatory pathways vary based on disease stage and the presence of SLE.

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INTRODUCTION

Chronic kidney disease (CKD) is a major public health issue that affects over 10% of the global population.[1] It is defined as a persistent reduction in kidney function with an estimated glomerular filtration rate (eGFR) below 60 mL/min/1.73 m^2 lasting for more than three months.[1] The kidneys are vital organs that are responsible for several functions, including filtration of waste products and excess fluid from the blood, maintaining acid-base balance, regulating blood pressure, production of various hormones, and stimulation of red blood cell production.[2] When kidney function declines, these processes are impaired, leading to various complications such as anemia, bone disease, electrolyte imbalance, metabolic acidosis, uremia, and edema.[3] CKD can progress to end-stage kidney disease (ESKD), which is the irreversible loss of kidney function that requires renal replacement therapy such as dialysis or transplantation.[4] CKD is also a major risk factor for cardiovascular disease (CVD), which is the leading cause of death among CKD patients.[5] Although ESKD represents the most advanced stage of CKD, this study makes distinctions between CKD and ESKD to enhance the clarity and interpretation of the findings. Systemic lupus erythematosus (SLE) is a complex autoimmune disorder that affects approximately five million people worldwide; it is characterized by the production of autoantibodies against various self-antigens, resulting in inflammation and damage to multiple organs and tissues.[6] SLE has a female predominance, with a female-to-male ratio of 9:1. It can affect individuals of any age group, but it predominantly manifests in those aged 15 to 45 years.[7] The clinical manifestations of SLE are heterogeneous and variable, depending on the organs involved and the severity of inflammation; some of the common symptoms include skin rash, arthritis, nephritis, serositis, hematologic, neurologic, and cardiovascular disorders.[8] SLE can impair kidney function due to lupus nephritis (LN), a serious condition affecting up to 60% of SLE patients.[9] SLE can also increase the risk of CVD by inducing endothelial dysfunction, atherosclerosis, and thrombosis.[10]

Inflammation and oxidative stress are key pathophysiological mechanisms involved in the development and progression of both SLE and CKD.[11] Inflammation is the body's response to injury or infection, which involves the activation of immune system, the release of inflammatory mediators, and the recruitment of inflammatory cells to the site of inflammation.[12] Inflammatory cytokines play an important role in mediating tissue injury and organ dysfunction in SLE and CKD, as well as in their complications.[13] Some of the most important inflammatory cytokines are interleukin-1β (IL-1β), interleukin-6 (IL-6), interleukin-12 (IL-12), and tumor necrosis factor-alpha (TNF-α). These cytokines play distinct roles in the pathogenesis of SLE and CKD. IL-1β and TNF-α are key mediators of acute inflammation, tissue injury, and immune cell recruitment; IL-6 exhibits both pro- and anti-inflammatory properties and is a potent driver of fibrotic tissue remodeling, also contributes to B cell differentiation and autoantibody production; while IL-12 promotes Th1 cell responses and enhances cytotoxic activity. Together, these cytokines drive immune activation, apoptosis, fibrosis, and vascular injury in SLE and CKD.[14,15]

The role of inflammatory cytokines in the pathogenesis and progression of CKD and SLE has been widely investigated, but the serum levels of these cytokines in CKD patients with or without SLE have not been adequately explored. Previous studies have reported conflicting results on serum IL-1β in SLE patients, depending on the measurement methods, the criteria of diagnosis, the stage of disease activity, and the presence of comorbidities.[16] Some studies have found no significant difference in serum IL-1β levels between SLE patients and healthy controls,[16] while others have identified higher levels of serum levels of IL-1β in SLE patients compared to controls.[17] Some studies have also reported lower levels of serum IL-1β in SLE patients with certain complications, such as immune thrombocytopenia.[18] Moreover, there is scarce data regarding serum levels of IL-6, IL-12, and TNF-α in patients with SLE with or without CKD. These cytokines are known to be involved in the regulation of immune responses, inflammation, and tissue damage in both CKD and SLE, and may have diagnostic and prognostic value for these conditions. The objective of this study was to compare the serum levels of IL-1β, IL-6, TNF-α, and IL-12 in CKD patients with o Inflammation and oxidative stress are key pathophysiological mechanisms involved in the development and progression of both SLE and CKD.[11] Inflammation is the body's response to

injury or infection, which involves the activation of immune system, the release of inflammatory mediators, and the recruitment of inflammatory cells to the site of inflammation.[12] Inflammatory cytokines play an important role in mediating tissue injury and organ dysfunction in SLE and CKD, as well as in their complications.[13] Some of the most important inflammatory cytokines are interleukin-1β (IL-1β), interleukin-6 (IL-6), interleukin-12 (IL-12), and tumor necrosis factor-alpha (TNF-α). These cytokines play distinct roles in the pathogenesis of SLE and CKD. IL-1β and TNF-α are key mediators of acute inflammation, tissue injury, and immune cell recruitment; IL-6 exhibits both pro- and anti-inflammatory properties and is a potent driver of fibrotic tissue remodeling, also contributes to B cell differentiation and autoantibody production; while IL-12 promotes Th1 cell responses and enhances cytotoxic activity. Together, these cytokines drive immune activation, apoptosis, fibrosis, and vascular injury in SLE and CKD.[14,15]

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MATERIALS AND METHODS

Study design

This study was an observational, cross-sectional study was conducted over a 15-month period in three governmental medical centers of Golestan Province, Iran (Sayyad Shirazi hospital, 5th Azar hospital, and Al-Jalil hospital) that included 118 participants, divided into three major groups based on their kidney function and SLE diagnosis: 38 CKD or ESKD patients with SLE (CKD/ESKD SLE+ group), 40 CKD or ESRD patients without SLE (CKD/ ESKD SLE- group), and 40 healthy controls (control group). The CKD or ESRD patients were recruited from the nephrology department of a tertiary hospital, and the healthy controls were recruited from the general population. The inclusion criteria for the CKD patients were as follows: age between 18 and 65 years, diagnosis of CKD stage 3 to 4 based on the estimated glomerular filtration rate (eGFR), and no history of renal replacement therapy. ESKD is defined as CKD stage 5 with an eGFR < 15 mL/min/1.73 m², typically requiring renal replacement therapy (dialysis or kidney transplant). To enhance study consistency and minimize confounding factors, we included only patients who were on a similar drug regimen or dialysis treatment protocol. The inclusion criteria for the healthy controls were: age between 18 and 65 years, normal renal function based on the eGFR, and no history of kidney or autoimmune disease. The classification of SLE was established based on the presence of at least four of the American College of Rheumatology criteria for SLE.[19] The exclusion criteria for all participants were: pregnancy, malignancy, infection, and other inflammatory conditions. Blood samples were collected from all participants after obtaining informed consent and following standard procedures. Serum samples were separated by centrifugation and stored at -80°C until analysis. Serum levels of IL-1β, IL-6, TNF-α, and IL-12 were measured using commercially available ELISA kits according to the manufacturer's instructions. The optical density was read at 450 nm using a microplate reader. The results were expressed as pg/ml.

Statistical analysis

The data were analyzed using SPSS software version 25. Descriptive statistics were used to summarize the demographic and clinical

characteristics of the participants. The differences in serum levels of cytokines among the three groups were compared using Kruskal-Wallis test, followed by the Mann-Whitney U test with Bonferroni correction for multiple comparisons. The normality of the data obtained from ELISA tests was evaluated using the Kolmogorov-Smirnov test. The analysis revealed that the data did not conform to a normal distribution pattern. Consequently, non-parametric statistical tests were utilized to assess and compare the findings of the study. A *P*-value ≤ .05 was considered statistically significant. Results are presented as median and interquartile range (IQR).

RESULTS

The main objective of this study was to compare the serum levels of pro-inflammatory cytokines in patients with chronic kidney disease (CKD or ESKD) with or without SLE and healthy controls. We also aimed to investigate the association between serum cytokine levels and the progression of CKD to ESKD. We measured the serum levels of IL-1β, IL-6, TNF-α and IL-12 using enzyme-linked immunosorbent assay (ELISA) in five groups of participants: CKD patients with SLE (CKD/SLE+), CKD patients without SLE (CKD/SLE-), ESKD patients with SLE (ESKD/SLE+), ESKD patients without SLE (ESKD/SLE-), and healthy controls.

Among the 31 patients with CKD two subsets were identified: those with systemic lupus erythematosus (SLE+; 58.1%), and those without (SLE-; 41.9%). Similarly, the 47 individuals with ESKD were divided into two subsets based on SLE positive (42.6%) and SLE negative (57.4%). Demographic and immunological characteristics of patients are summarized in Table 1.

Comparison of serum cytokine levels based on SLE status

Serum cytokine levels were compared across the three main study groups. A significant difference was found in the serum levels of IL-1β, IL-6, and IL-12 using the Kruskal-Wallis test, as depicted in Figure 1. Specifically, serum levels of IL-1β were significantly elevated in CKD/ESKD SLE+ patients compared to the both CKD/ESKD SLE- and control groups (Figure 1A).

IL-12 levels were found to be lower in the CKD/ESKD SLE+ group compared to the other two groups, although these differences were significant in comparison to the control group (Figure 1B). CKD/ESKD SLE- patients exhibited remarkably higher TNF-α in their serum compared with the control individuals (Figure 1C).

IL-6 concentrations were significantly higher in both patients with and without SLE groups than in healthy individuals. Besides, among these patients, the SLE- group had relatively higher IL-6 than the SLE+ patients (Figure 1D). Median (inter-quartile) of different groups of the study are presented in the Table 2.

Table 1. Demographic and immunological characteristics of patients (presented in median, IQR)

Parameters	ESKD (N=47)	CKD (N=31)	ESKD/SLE+ (N=20)	CKD/SLE+ (N=18)
Age at the study time (year)	53 (44, 62)	49 (39, 67)	47 (37.5, 58)	54 (38.50, 67.50)
Sex ratio (F/M) (%)	31/16 (66%/34%)	21/10 (67.7%/32.3%)	17/3 (85%/15%)	13/5 (72.2%/27.8%)
WBC (cells/mm^3)	9.4 (7.9, 10.1)	9.2 (7.9, 10)	9.20 (7.83, 10.08)	9.30 (7.80, 10.30)
RBC (cells/mm^3)	3.44 (3.18, 4.07)	3.57 (3.27, 4.05)	3.71 (3.16, 4.08)	3.44 (3.15, 3.78)
Neutrophils; (cells/mm^3)	59.50 (55.00, 67.75)	61.00 (55.00, 70.00)	64.00 (58.75, 77.75)	63.50 (54.25,69.00)
Lymphocytes; (cells/mm^3)	18 (11, 22)	19 (12, 23)	17.00 (10.25, 22.75)	18 (11.75, 23.50)
Hemoglobin; g/dl	10.10 (9, 12)	9.5 (9, 11)	11.15 (9.75, 12.30)	9.40 (8.90, 10.63)
PLT; (cells/mm^3)	169 (123, 232)	211 (132, 293)	176.5 (128.5, 243.5)	207 (141, 299.50)
BUN (mg/dL)	112.5 (98.75, 129.75)	89 (76, 99)	113 (100, 144)	91 (78, 104)
Cr (mg/dL)	7.60 (6.02, 9.60)	3 (2,4)	7.30 (3.22, 9.20)	3.30 (1.75, 4.80)
Uric Acid (mg/dL)	5.50 (4.80, 7.40)	6 (5, 7.5)	5.50 (4.85, 7.35)	6.30 (4.40, 7.58)
Total Protein (g/dL)	6.4 (5.8, 6.9)	6.80 (5.93, 7.23)	6.55 (5.63, 7.10)	6.60 (5.55, 7)
Albumin (g/dL)	3.80 (3.60, 4.05)	3.40 (2.98, 3.95)	3.80 (3.60, 4.10)	3.50 (2.98, 3.95)
ESR (mm/h)	43 (25.75, 54)	49 (45, 67)	38.50 (24.50, 52.50)	53.5 (47.25, 67.50)

BUN: blood urea nitrogen; CKD; chronic kidney disease; CPK: Creatine Phosphokinase; ESR: Erythrocyte sedimentation rate; ESKD; End-Stage Kidney Disease, M: Male; F: Female; PLT: platelet; PTH: parathyroid hormone; RBC: red blood cell; WBC: white blood cells. IQR: range with 75th and 25th percentiles.

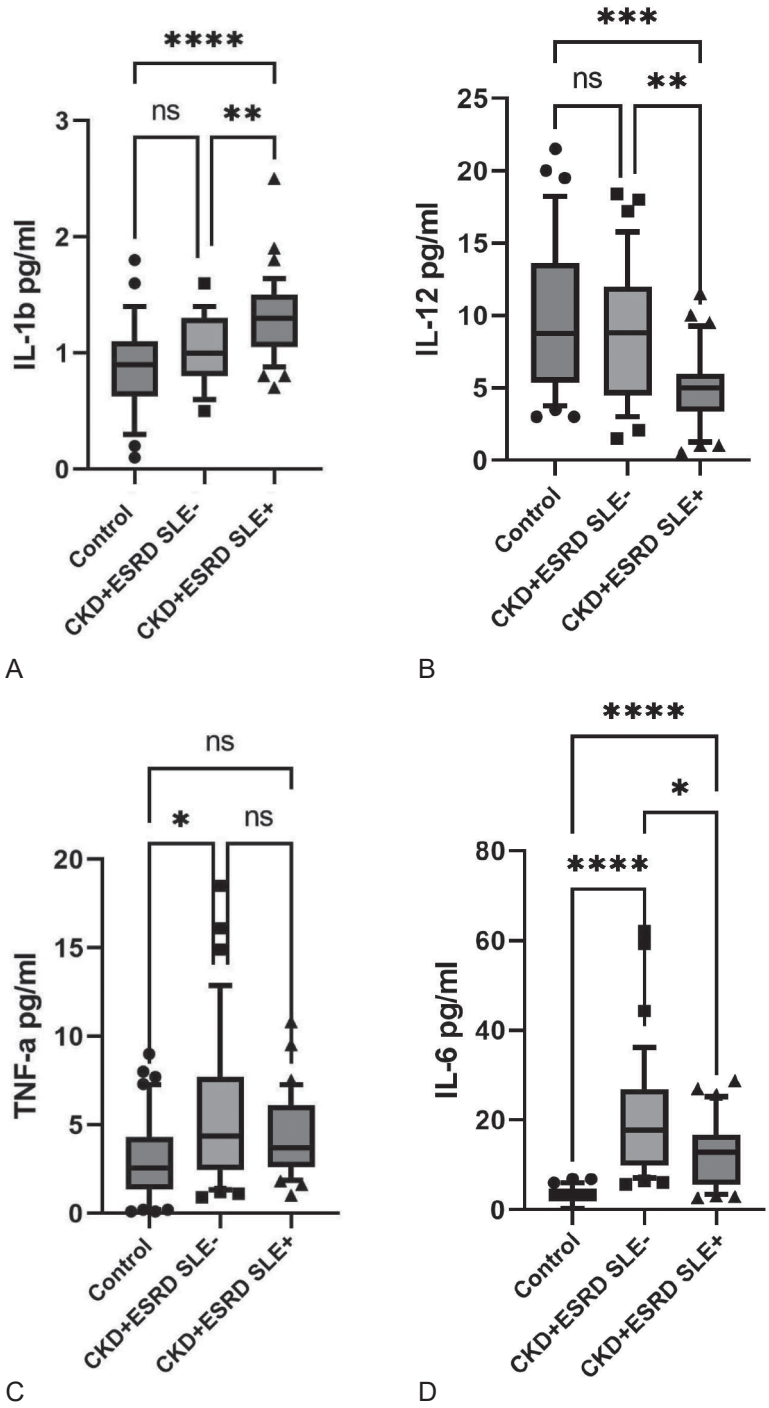

Figure 1. Comparison of serum IL-1β (A), IL-12 (B), TNF-α (C) and IL-6 (D), between the CKD/ESKD SLE-, CKD/ESKD SLE+ and controls. * < 0.05, ** < 0.01, *** < 0.001, **** < 0.0001, ns: non-significant.

Table 2. Comparison of serum cytokine levels based on SLE status

Variables	SLE Positive Median (inter-quartile)	SLE Negative Median (inter-quartile)	Control Median (inter-quartile)	*P*
IL-1β (pg/mL)	1.3 (1.0, 1.5)	1.0 (0.72, 1.3)	0.9 (.6, 1.2)	.001
IL-12 (pg/mL)	5.0 (3.8, 8.5)	8.0 (4.4, 12.9)	8.7 (5.0, 14.7)	.023
TNFα (pg/mL)	3.5 (2.1, 6.0)	4.1(1.8, 7.7)	2.5 (1.3, 4.2)	.075
IL-6 (pg/mL)	13 (5.5, 18.2)	16.0 (8.9, 26.9)	3.9 (1.8, 5.8)	0 >.001

Comparison of serum cytokine levels between CKD and ESKD patients

The expression of the inflammatory cytokines was compared between different CKD stages regardless of SLE status; using the non-parametric Mann-Whitney U test (Figure 2, Table 3). The analysis revealed that among the tested cytokines, both IL-12 and TNF-α were significantly increased in the ESKD group (Figure 2B, C).

In comparison between the CKD patients and the

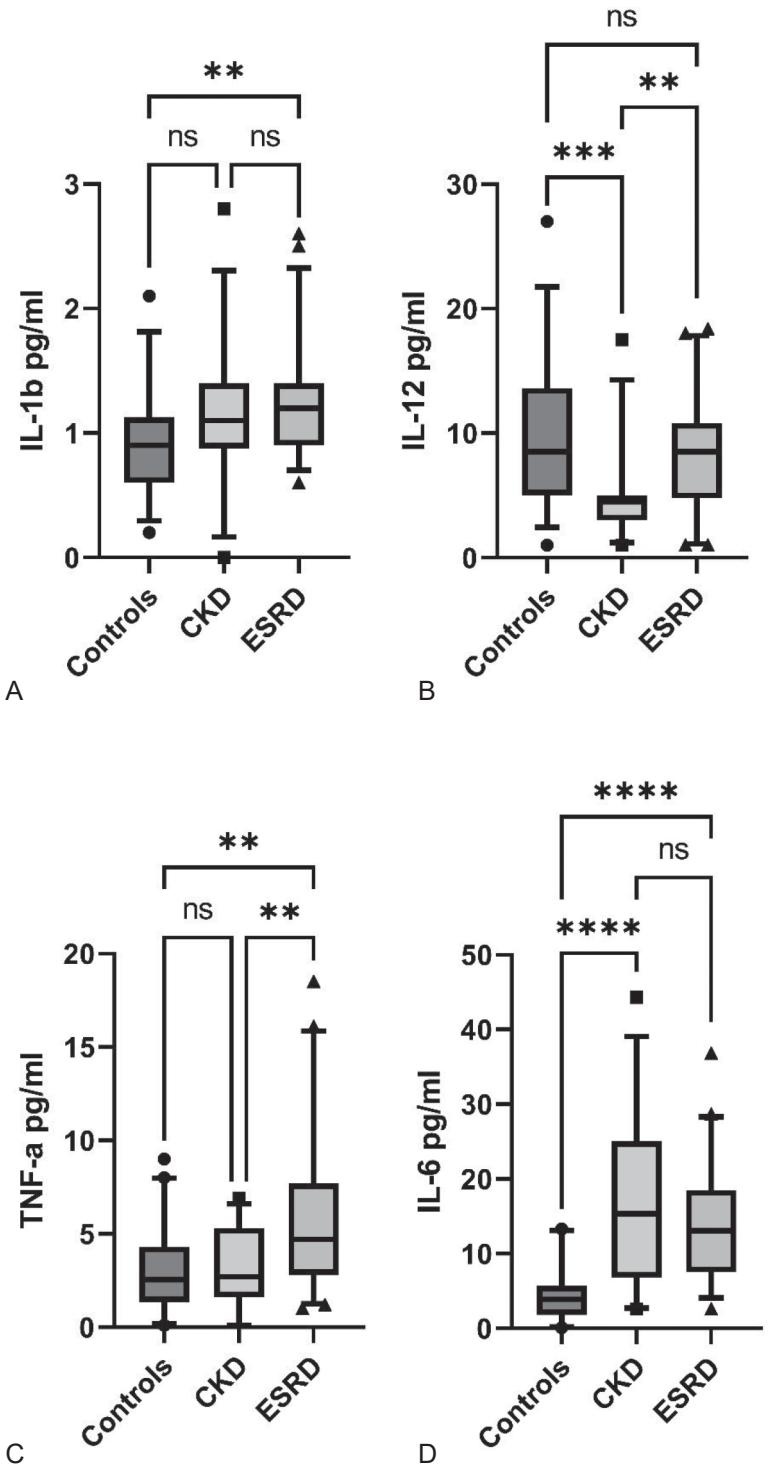

Figure 2. Comparison of serum IL-1β (A), IL-12 (B), TNF-α (C) and IL-6 (D), between the CKD and ESKD patients and the control group. * < 0.05, ** < 0.01, *** < 0.001, **** < 0.0001, ns: non-significant.

controls, it was observed that IL-6 was significantly higher in CKD patients (Figure 2D), whereas the concentration of IL-12 was significantly lower in patients (Figure 2B).

A comparison between the ESKD patients and the controls, revealed that IL-1β, IL-6, and TNF-α were significantly higher in ESRD patients (Figure 2 A, C, D).

Comparison of the cytokine levels between CKD and ESKD patients with or without SLE.

As summarized in Tables 4, 5 and Figure 3; serum levels of IL-1β, IL-12, TNF-α and IL-6 were compared between the study groups using the non-parametric Mann-Whitney U test. IL-6 was increased in the ESKD/SLE+ group compared with CKD/SLE+, though not statistically significant (Figure 3D).

On the other hand, when comparing the levels of the pro-inflammatory cytokines between CKD/SLE- and ESKD/SLE- patients, the serum concentrations of IL-1β, IL-12, and TNF-α were significantly increased in ESKD patients (Figure 3 A-C); whereas, it was observed that IL-6 concentration was significantly lower in ESKD patients (Figure 3D).

Significant reduction of IL-12 and TNF-α in ESKD/SLE+ patients in comparison to ESKD/SLE- patients was observed (Figure 3B, C), Table 4.

Also, IL-1β, IL-12, and TNF-α were significantly increased in CKD/SLE+ patients compared to the CKD/SLE- patients; furthermore, IL-6 was significantly elevated in CKD/SLE- patients compared to the CKD/SLE+ patients (Figure 3D), Table 5.

DISCUSSION

The current study revealed that the serum levels of IL-1β, IL-12, TNF-α, and IL-6 were considerably altered in patients with kidney diseases with or without SLE. These findings suggest that the inflammatory cytokine profile is influenced by the presence SLE and the stage of kidney disease. First, we compared the serum levels of the inflammatory cytokines between the CKD/ESKD patients with or without SLE; we found that IL-1β was significantly higher in SLE+ patients, while IL-6 was significantly lower among this subset of patients. The lower serum concentration of IL-6 among SLE+ patients might be due to the heavier disease-modifying treatments that could modulate IL-6, or it may have a protective effect in the CKD/ESKD SLE- patients. Similarly, in comparison

Table 3. Comparison of serum cytokine levels between CKD and ESKD patients

Variables	CKD Median (inter-quartile)	ESRD Median (inter-quartile)	Control Median (inter-quartile)	*P*-value CKD & ESRD	*P*-value CKD & Control	*P*-value ESRD & Control
IL-1β (pg/mL)	1.1 (0.8, 1.4)	1.3 (0.9, 1.5)	0.9 (.6, 1.2)	.178	.1490	.002
IL-12 (pg/mL)	4.5 (3.0, 5.0)	8.8 (4.8, 12.0)	8.7 (5.0, 14.7)	.001	< .001	.520
TNFα (pg/mL)	2.7 (1.5, 5.2)	4.6 (2.7, 7.7)	2.5 (1.3, 4.2)	.003	.817	.002
IL-6 (pg/mL)	16.6 (6.7, 27.4)	14.1 (8.1, 21)	3.9 (1.8, 5.8)	.650	<.001	< .001

Table 4. Comparison of Cytokine Levels between ESRD Patients with and without SLE

Variables	ESRD & SLE. Positive Median (inter-quartile)	ESRD & SLE. Negative Median (inter-quartile)	Asymp. Sig. (2-tailed)
IL-1β (pg/mL)	1.3 (0.9, 1.5)	1.1 (0.9, 1.4)	0.138
IL-12 (pg/mL)	5.0 (3.3, 7.8)	9.6 (7.2, 14)	< 0.001
TNFα (pg/mL)	3.2 (1.9, 6.5)	5.8 (3.6, 8.9)	0.020
IL-6 (pg/mL)	14.5 (10.8, 25.4)	11 (7.3, 18.9)	0.263

Table 5. Comparison of Cytokine Levels between CKD Patients with and without SLE

Variables	CKD & SLE. Positive Median (inter-quartile)	CKD & SLE. Negative Median (inter-quartile)	Exact Sig. [2*(1-tailed Sig.)]
IL-1β (pg/mL)	1.2 (1.0, 1.4)	0.9 (0.4, 1.0)	0.003
IL-12 (pg/mL)	5.0 (3.8, 8.7)	3.5 (1.8, 4.5)	0.006
TNFα (pg/mL)	4.0 (2.5, 5.8)	1.7 (0.2, 2.8)	0.006
IL-6 (pg/mL)	6.9 (3.6, 14.7)	27.4 (20.2, 31)	< 0.001

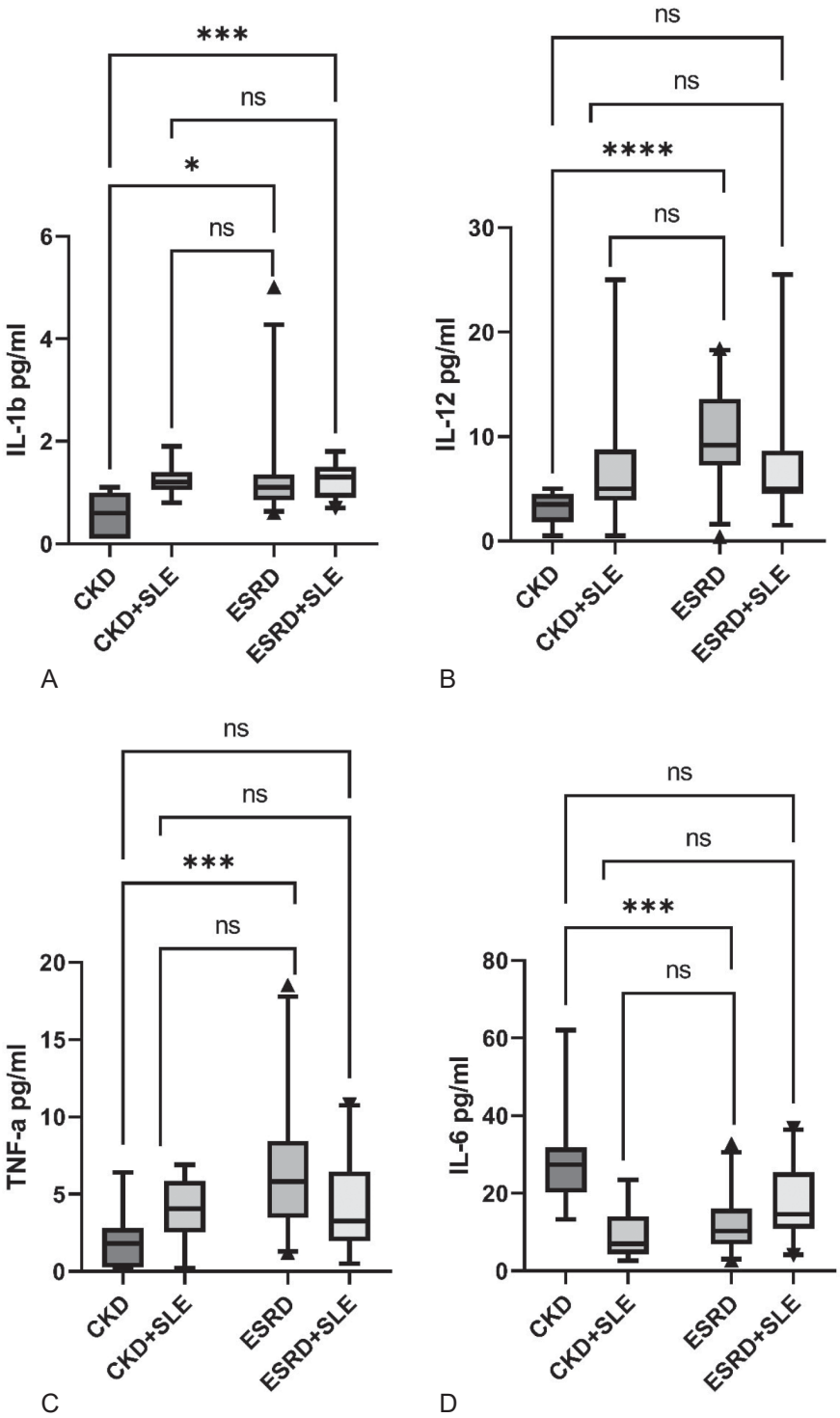

Figure 3. Comparison of the IL-1β (A), IL-12 (B), TNF-α (C), and IL-6 (D) concentrations between the CKD and ESKD patients within the SLE- and SLE+ cohorts. * < 0.05, ** < 0.01, *** < 0.001, **** < 0.0001, ns: non-significant.

between the CKD/ESKD SLE+ patients and the control group, it was revealed that both IL1β and IL-6 were significantly higher in SLE+ patients, while IL-12 was significantly lower among these patients. Data around the IL-1β levels in SLE patients are mixed, and they do not demonstrate a consistent pattern;[18,20,21] for example, Mende *et al.* have demonstrated that serum IL-1β in patients with

SLE was not significantly different from healthy individuals.[16] Whereas, in an individual study by Wu *et al.*, it was reported that both IL-1β and IL-6 were significantly increased in sera from patients with SLE compared to healthy individuals,[22] which is consistent with our results showing elevation of IL-1β and IL-6 in sera from CKD/ESKD SLE+ patients. Regarding the IL-6 serum levels in SLE patients, multiple studies have reported increased IL-6 in patients' sera which is in line with our data showing significant increase in IL-6 levels in patients compared to the controls.[23-25] For instance, in a meta-analysis consist of 24 studies by Ding *et al.*, showed that IL-6 was significantly higher in sera from patients with SLE in comparison to healthy individuals and the IL-6 levels were significantly correlated with disease severity.[26] Consistent with our data showing a significant reduction of IL-12 in CKD/ESKD SLE+ patients, in two separate but related pioneer studies by Liu *et al.*, reported that the IL-12 expression in SLE patients is downregulated both at mRNA and protein levels and is negatively correlated with serum IL-10.[27,28] Conversely, Tokano *et al.*, and Li *et al.*, reported that IL-12 was significantly increased in SLE patients' sera in comparison to healthy individuals, which is in contrast to our findings.[29,30]

Furthermore, the profile of inflammatory cytokines was compared between the CKD and ESKD patients, irrespective of the presence of SLE, and the healthy controls; it was observed that both TNF-α and IL-12 were significantly increased in ESKD patients compared to patients with CKD, suggesting a higher inflammatory profile among this subset of patients. On the other hand, the with CKD patients had significantly higher IL-6 compared to the controls, while they had lower concentrations of IL-12. The rise in IL-6 levels among patients affected with different inflammatory condition is an indisputable event, as this cytokine is promptly upregulated under physiologic disturbances related to inflammation and exerts both local and systemic effects on the immune system.[31] Lower IL-12 in CKD patients compared to the controls could be explained by the notion that IL-12 could be explained by its protective effects against renal injury through immune modulation and reduction of fibrotic.[32-34] Therefore, the imbalance between IL-6 and IL-12 in CKD patients may reflect a dysregulated immune system that favors inflammation and tissue damage. We also observed that IL-1β, TNF-α and IL-6 were significantly higher in ESKD patients compared to healthy controls. This could be explained by the fact that advanced stages of CKD and ESKD are associated with a heightened inflammatory profile due to various factors contributing, such as accumulation of detrimental substances.[35] Lisowska *et al.* showed elevated levels of IL-6 in CKD patients compared to the controls which is quite consistent with our data.[36] In consistent with our data, in another randomized clinical trial it was observed that IL-6 was increased in CKD patients.[37]

In further analyses, patients with CKD/SLE+ were compared with those with ESKD/SLE+, revealed that only IL-6 was significantly higher in ESKD/SLE+ patients. Comparison between the CKD/SLE- and ESKD/SLE- patients showed that IL-1β, IL-12, and TNF-α were significantly higher in ESKD/SLE- patients, while IL-6 was significantly higher in CKD/SLE- patients. These results indicate that IL-6 may be one of the cytokines implicated in the pathogenesis of kidney disease, especially in patients with SLE. IL-6 can promote inflammation, fibrosis, and immune cell activation in the kidneys and may also reflect the severity of kidney damage and inflammation in SLE patients. On the other hand, significantly higher IL-1β, IL-12, and TNF-α in ESKD/ SLE- patients, and significantly higher IL-6 in CKD/SLE- patients suggest that these cytokines may have different roles and mechanisms in the development of kidney disease without SLE. IL-1β, IL-12, and TNF-α levels may be more affected in the advanced stages of kidney disease, while IL-6 may be more affected in the early stage. These results indicate that IL-6 is a common and crucial factor in kidney disease, but its role may vary depending on the presence or absence of SLE. In this regard, targeting IL-6 could be a potential therapeutic strategy for kidney disease, especially for patients with SLE. Perlman *et al.* reported that IL-6 level was elevated in patients with diabetic nephropathy at earlier stages of CKD, but remained unchanged at ESKD.[38] Although IL-6 is deemed a pro-inflammatory cytokine, it can also have anti-inflammatory effects by stimulating the production of IL-10 and IL-1 receptor antagonists; it is also involved in the regulation of B cell differentiation and antibody production, which are important in the pathogenesis of SLE.[39,40] Besides, the lower

levels of IL-6 in SLE+ patients may be due to several factors; one possibility is that IL-6 is consumed by binding to its receptors on various cells, such as B cells, T cells, hepatocytes, and endothelial cells. Another possibility is that IL-6 is downregulated by negative feedback mechanisms, such as the induction of suppressor of cytokine signaling (SOCS) proteins or the activation of glucocorticoid receptors. A third possibility is that IL-6 is counteracted by other cytokines or factors that inhibit its signaling or expression, such as IL-10, IL-1 receptor antagonist, or transforming growth factor-beta (TGF-beta).[41-43]

Kir *et al.*, previously demonstrated that TNF-α levels are significantly increased in patients with CKD in comparison with the healthy individuals, besides they also showed that the TNF-α levels are higher in patients undergoing dialysis compared to these who are pre-dialysis,[44] suggesting that the TNF-α levels are increased with the disease progression. In an animal study using an MRL-lpr mouse, Schwarting *et al.* have demonstrated that tubular cells within the kidney are capable of secreting IL-12 and its forced overexpression may induce the infiltration of various T cell subsets to the kidney, which leads to the inflammatory kidney damage.[45] This partly explains why individuals affected with autoimmune and inflammatory disorders such as SLE are more prone to developing CKD. One underlying mechanisms for this predisposition could be elevated levels of IL-12, which was observed to be higher among CKD/SLE+ patients. Therefore, monitoring and modulating the levels of these cytokines may be beneficial for managing kidney disease in SLE+ patients.

The study found that in the CKD subgroup, IL-1β levels were significantly correlated with TNF-α levels, which could suggest that these molecules are involved in the pathogenesis or progression of CKD, or that they reflect the severity of kidney damage. Some studies have suggested that IL-1β, TNF-α and C-C motif chemokine ligand 5 (CCL5) could modulate the inflammatory and fibrotic processes, therefore they may play a role in the pathogenesis or progression of CKD as well.[46] For example, IL-1β and TNF-α can induce the expression of pro-fibrotic factors, such as TGF-b and collagen, in renal cells. They can also activate the NF-κB pathway, which regulates the expression of various inflammatory genes, such as CCL5.[47-50] Therefore, IL-1β, TNF-α and CCL5 may have significant roles in CKD as potential biomarkers or therapeutic targets. However, more studies are needed to elucidate the exact mechanisms and interactions of these markers in CKD, as well as their effects on other organs or systems.

LIMITATIONS OF THE STUDY ARE AS FOLLOWS

The heterogeneity of the patient groups may introduce confounding factors that influence the levels of immunomodulatory factors, such as age, sex, ethnicity, comorbidities, medications, treatments, or interventions. Therefore, further studies with larger and more homogeneous samples are needed to confirm and extend these findings. Moreover, this study assessed only the serum levels of immunomodulatory factors, which may not reflect their local expression or activity in the kidney or other organs. Thus, further studies using tissue or urine samples are needed to explore the molecular mechanisms and pathways of these factors in the pathogenesis and progression of CKD and SLE.

CONCLUSION

The findings underscore the critical role of IL-6 in the pathogenesis of kidney disease, particularly in patients with SLE. Elevated IL-6 levels were observed consistently in patients with CKD compared to those with ESKD, regardless of SLE status, and were strongly associated with the severity of kidney damage in patients with SLE, suggesting its central role in mediating inflammation and kidney injury. In contrast, IL-1β, IL-12, and TNF-α appeared to variable pattern, indicating that their mechanisms of action may differ depending on disease stage and the presence or absence of SLE. Future studies should explore the molecular mechanisms underlying these cytokine-driven processes and evaluate their therapeutic implications of IL-6 for kidney diseases in clinical settings.

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CONFLICTS OF INTEREST

The authors declared no conflict of interests.

Ethical considerations

The research conducted in this study adhered to the principles outlined in the Declaration of Helsinki and was approved by the Ethics Committee of Golestan University of Medical Sciences (Ethical code No. IR.GOUMS.REC.1402.099). Prior to any intervention, all participants provided written informed consent. The authors have fully complied with ethical issues, such as plagiarism, data fabrication, and double publication.

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