The Immunological Role of Interleukin-10 in Chronic Renal Disease Patients of Thi-Qar Province

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Abstract-: Chronic renal disease (CRD) is a basic worldwide health issue; the interleukin-10 is an anti-inflammatory mediator that has a role in normal kidney function and in the progression of chronic renal failure. This study aims to measure the serum level of anti-inflammatory cytokines (IL-10), and evaluate its role in the progression of chronic kidney disease (CRD). The study comprised 66 patients divided into group 1 (22 subjects with CKD) and 2 (22 CRD patients with Diabetic mellitus name as DM-CRD), and group 3 (22 patients with Diabetic mellitus DM only without CRD). In addition to 22 consider as healthy people as control. The serum levels of IL-10 were determined by using ELISA assay. The results demonstrated a significant increase in the serum means levels of IL-10 in all patient groups (CKD, DM-CRD, DM) in comparison with healthy control, while there was nonsignificant difference in its mean serum levels among all patient groups. It was concluded that male and aging are predisposing factors to chronic renal disease, and elevated levels of IL-10 in patients with CKD and diabetes reflect the potential role of IL-10 in the progression of diabetic nephropathy patients to chronic renal disease .

Keywords: IL-10, Anti-Inflammatory cytokine, CRD, Renal function tests.

I. INTRODUCTION

Kidney diseases such as chronic kidney disease (CKD) and diabetic nephropathy (DN) are a world health issue with raised mortality and economic burden [1]. IL-10 is an anti-inflammatory cytokine that keeps tissue homeostasis by inhibiting excessive inflammatory response, regulating immunosuppressant, and delaying tissue fibrosis, enhancing tissue repair during severe infection in kidney disease. So, improving the pathomechanism and identifying effective therapeutic targets is significant for the treatment and diagnosis of kidney disease [1].

One of the most devastating microvascular complications of diabetes in approximately one-third of patients is diabetic kidney disease (DKD), which currently represents the leading cause of end-stage renal disease (ESRD) [2].

Inflammation is the body's natural and necessary response to various stimuli. It allows immune system cells to move to the target site of stimuli through a series of steps that are coordinated by cytokines, chemokines, and acute-phase proteins. In mild cases, this provides a solution to the problem and allows a return to homeostasis. On the other hand, chronic inflammation can contribute to tissue damage and fibrosis. Therefore, it has been linked to many diseases, including chronic kidney disease [3].

Interleukin-10 (IL-10) accounts as an anti-inflammatory cytokine which produced by activated immune cells and has a critical and significant role in immune responses control [4]. A previous study by Sinuani and his colleagues indicated that IL-10-induced signaling pathways serve an important role in regulating and maintaining normal kidney function [5].

Furthermore, Yilmaz et al. found that higher sera levels of IL-10 presented in patients with low glomerular filtration rate eGFR in in addition to other inflammatory markers [6], another study indicated that adverse cardiovascular events in chronic kidney disease associate with circulating interleukin (IL)-10 [7] .

Therefore, Considering the aforementioned literature about the importance of interleukin 10 and its role in kidney disease, the current study was designed to measure the level of this anti-inflammatory cytokine in patient samples and healthy controls and evaluate its role in CRD progression.

II. PATIENTS AND METHODS

A. Patients and Control

This study was conducted in the Department of pathological analysis in Thi_Qar University, College of Science.

The current study enrolled 44 chronic renal failure patients of both sexes who visited the Al-Hussein Dialysis Center in the Imam Al-Hussein Teaching Hospital in Thi Qar Governorate, southern Iraq, and 22 diabetic patients (positive control) who visited the Diabetes and Endocrinology Center in Thi Qar between October 15, 2023, and January 31, 2024. In addition to 22 healthy people. Their Ages ranged (29-72). Patients are diagnosed based on biochemical examination, in addition to the role of physicians in confirming the diagnosis. Chronic renal

disease patients are classified into two groups based on whether they have diabetes or not.

B. Method

Three milliliters of whole venous blood samples were drawn from CRD patients (pre-dialysis session), DM patients, and healthy controls, then they were placed in gel tubes and left to clot at room temperature, to get serum after separate it by centrifuged to at 4000 xg for 10 min. The serum of the gel tube was put into an Eppendorf tube and stored at (-20℃) until used to measure creatinine, and urea by the enzymatic colorimetric method by using multi chemical fully automated chemistry analyzer (Abbott/USA) and to measure sera levels of IL -10, by a sandwich human IL-10 ELISA kit according to the instructions of the supplier Sunlong Biotech Co., LTD Company – China Cat No. CSL0967Hu.

C. Statistics Analysis

The data analyzed using SPSS version 26. Percentages were used with categorical variables such as age and gender. The results presented as a mean ± standard deviation (SD). p-values ≤ 0.05 were interpreted as significant. Chi-square and one-way ANOVA were used to analyze the results of this study .

III. RESULTS

A. Demographical Characteristics

Distribution of the subject groups according to gender, is shown in Figure 1, where the results indicated that the predominant of CRF patients were males, with a percentage of 26/44 (59.1%) males, while the percentage of females was 18/44 (40.9%), and the largest percentage appeared for males as well in diabetic patients, it was 14 (63.6%) males, and only 8 (36.4%) were females, while the number of males and females was equal in the healthy group at 11 (50%) for each. It became clear from the results of the current study males are more frequent in all groups of patients, as shown in Figure (1). Males to females was 1.3:1, with a slight predominance of males over females in chronic kidney disease, There were non-significant differences when compared among studied groups according to gender among all studied groups $(x^2 = 0.829, P \text{ value} = 0.886)$.
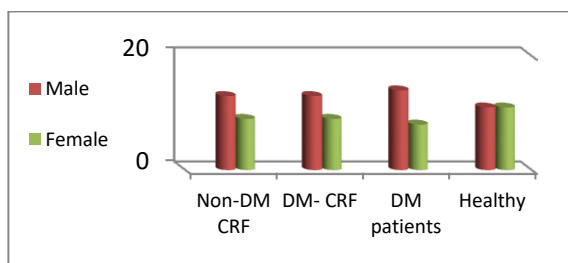

Figure 1: Distribution of patients and healthy participants according to sex

Regarding age, a total of 88 participated were involved in this study , among them were 44 CKD patients (ages range 29-72 , mean age 56.8± 14.02) , 22 of them were DM patients(ages range 29-72 , mean age mean 52.6±10.5) , in addition to 22 healthy people of (ages range 29-63, mean

age mean 42.7± 10.4) . Moreover, the results showed there was a significant difference(X^2=23.939, P- value= 0.004) in the distribution of patients and healthy according to age groups as the largest percentage of elderly people were more likely to suffer from kidney failure, and the maximum number of chronic kidney disease patients in both groups (CKD with and without Diabetic) was within the age group 62-72 years 21/44 (47.73%). The distribution of the remaining cases of the study patients and healthy controls within different age groups is listed in Figure (2).
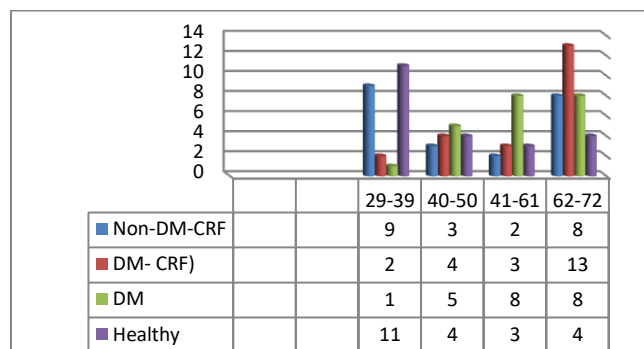

	29-39	40-50	41-61	62-72
Non-DM-CRF	9	3	2	8
DM- CRF)	2	4	3	13
DM	1	5	8	8
Healthy	11	4	3	4

Figure (2): Distribution of studied subjects regarding Age groups

B. Serum Levels of Interleukin 10 (IL-10)

ANOVA test was used to analyze the present results of serum IL-10 levels, where the results indicated a significant difference in IL-10 concentration among study groups (F=37.618, P<0.01). The present data also showed a significant increase in the mean serum IL-10 level in all patient groups (CRF, DCRF, DM) compared to the healthy control group (31.328 pg/mL, 33.851 pg/mL, and 34.052, respectively) compared to (10.049 pg/ml) in the control group, while the results did not show any significant difference between the patient groups as referred in Table (1).

Table (1): Serum levels of IL-10 in patient and healthy

Groups	Mean± S.E. of IL-10 pg/ml	Sig. between groups	P value
CRD	31.329±8.593	CRF X DCRF	0.297 [NS]
DM- CRD	33.851±7.852	CRF X DM	0.260 [NS]
DM	34.052±10.716	DM X DCRF	0.933 [NS]
Healthy	10.049 ± 1.361	CRF X Healthy	0.00**
		DM X Healthy , DCRF X Healthy	0.00**

The current data also showed significant an elevated mean serum levels of urea, creatinine in both groups of CRD patients with and with no DM (CRF and DM-CRF) compare to healthy groups and only DM patients where (urea; 149.82±43.88, 130.03±41.42; creatinine;8.29±2.42, 7.22±2.52[c] ; as compared with the healthy control and DM, respectively (urea; 23.64±7.49, 31.73±10.24; creatinine; 0.83±0.16, 1.003±0.17;. while, there was no significant difference between control groups (healthy and DM) .

Table 2 :Comparison of urea and creatinine in patients and healthy volunteers

Parameters	Urea(mg/dl)	Creatinine(mg/dl)
Healthy	23.64±7.49[a]	0.83±0.16[a]
CRF	149.82±43.88[b]	8.29±2.42[b]
DM-CRF	130.03±41.42[c]	7.22±2.52[c]
DM	31.73±10.24[ad]	1.003±0.17[a]
P value	0.00[**]	0.00[**]

Table(3) :Correlation coefficient between IL-10 renal function parameters

Person correlation		Urea	Creatinine
IL-10	P. value	0.07	0.357[*]
	r. value	0.6	0.02

IV. DISCUSSION

The current findings showed that more than half of the patients with chronic kidney failure were males (59.1%) and their ratio to females was 1.3:1. This result is consistent with a previous local study in Thi-Qar, which indicated that kidney failure is more common in men than in women, and the ratio of males to females was 1.6:1 [8].

Also, the results in line with Iraqi study conducted in Baghdad found that male proportion among CRF Patients was (54.7%) [9]. Moreover, Muhammad-Baqir *et al.* revealed that male patients represented the largest proportion of kidney failure patients at 52.5% [10].
Kidney disease occurs more frequently in males than females, and is due to differences in testosterone, which in a sex-specific manner affects kidney function and chronic kidney disease and, it is the underlying cause of chronic kidney disease in males but not in females [11].
Moreover, estrogen, which is abundant in women until menopause, plays a role in protecting women's kidneys [12]
On the other hand, the present findings showed that kidney failure was more occur in the elderly people, and the maximum number of kidney failure patients was 21 patients at 47.73% and within the age group of 62-72 years. This result is consistent with [9] and [8] which found that the highest incidences of CRF patients were within elderly subjects.

As shown in Table (1), the results indicated there was a significant increase of IL-10 in all chronic renal patients with DM compare with healthy groups. The current results were in line with Iraqi study by Azeez and Darogha who indicated there was a significantly an increased levels of IL-10 (26.29 ± 2.422) in chronic renal patients on hemodialysis when compared with control group (15.00 ± 0.918) [13] .The results also are in agreement with those of the study carried out by Saleh and Ahmed in Baghdad city, who reported that the mean concentration of IL-10 was significantly increased above normal level for all ESRD

patient's groups (Hypertensive, Diabetic, both hypertensive and Diabetic) subjected to hemodialysis [14] . Published data has revealed that the serum level of IL-10 increases as a result of uraemic monocytes and that kidney clearance drops in ESRD[13]. Myśliwska et al. found that diabetic nephropathy patients had elevated levels of circulating interleukin-10 [15] .
The current results were dis-agree with a study in Iraq by Shyeaa *et al.* who indicated there was a significantly a decreased levels of IL-10 (26.20 ±5.16) in chronic renal patients when compared with control group (79.27 ±23.44) [16].

IL-10 has a significant role in normal kidney physiology, and during acute kidney injury and in the progression of chronic renal failure. Mesangial cells are also the major local source of IL-10 in normal adult kidneys and represent key regulators of renal function [5].

The present findings also recorded a significant increase in the levels of IL-10 in the blood serum of diabetic patients only without CKD compared to healthy controls. While the results did not indicate any significant difference in IL-10 levels among chronic renal failure patients with diabetes compared to patients with diabetes only, with a slight increase in IL-10 levels in DM patients compare with DM-CKD. This explains the key role that IL-10 plays in chronic renal failure development and outcomes and it may be useful as an early marker of kidney dysfunction in patients with DM type II.

Generally, the results of this study are agreement with previous reports showing an association between IL-10 regulation and the pathophysiology of various kidney diseases, such as diabetic nephropathy, where elevated serum IL-10 can predict albuminuria and is associated with nephropathy severity [5,15] .

The circulating IL-10 level helps cells combat the chronic low-grade inflammation in obesity and T2DM. In contrast, low IL-10 level indicates that the regulatory mechanism has failed, resulting in the activation of monocytes and lymphocytes [17].

In the current study, the mean of the urea and creatinine levels showed high significant increase in patients with CKD compared with healthy individuals (p ≤ 0.01) (Table 2). These results agree with resent Iraqi study [18] and other study [19] The explanation for this increase in the levels of urea and creatinine is due to the decrease in the number of nephrons in patients with kidney failure, which leads to a reduced in the glomerular filtration rate (GFR), which in turn causes a significant decrease in the level of water and solutes. Ultimately, the kidney loses its efficiency in removing nitrogenous wastes from the blood, which leads to the accumulation of these wastes in the bloodstream [20] [21]. Finally, the results showed that there is a positive correlation between higher IL-10 levels and higher blood urea and creatinine levels in patients with chronic kidney disease. Therefore, this may indicate that IL-10 may reflect the degree of renal dysfunction and the severity of renal impairment.

V. CONCLUSSIONS

It was concluded that male, aging are predisposing factors to chronic renal disease. An elevated mean serum levels of IL-10 in chronic kidney disease patients and diabetes

patients reflect the potential role of IL-10 in the progression of diabetic nephropathy to chronic renal disease. So, it is considered as an indicator sign of decreased kidney function. Moreover, the measurement of IL-10 and conventional renal biomarkers holds promise for improving clinical management and outcomes in patients with chronic kidney disease .

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

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