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DIAGNOSTIC VALUE OF INTERLUKEIN-18 IN RENAL FAILURE DISEASE

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ABSTRACT

Background: End-stage renal disease (ESRD) is the last stage of chronic kidney disease which is related with significantly increased morbidity and mortality. Interleukin-18 (IL-18), a strong proinflammatory cytokine with multi properties. The aim was to evaluate the role of pro-inflammatory cytokine Interleukin-18 in end stage renal disease. **Methods:** This study was conducted on 150 subjects all collected from Benha teaching Hospital and were divided into three groups Group (1): control group composed of 50 healthy volunteer individuals. Group (2): 50 patients with renal failure. Group (3): 50 patients with renal failure and inflammation. Serum IL-18 was measured for all subjects. **Results:** Mean of IL-18 in controls, patients with renal failure, patients with renal failure and inflammation were 98.50±60.01, 548.78±140.73 and 1114.16±243.03 respectively. Mean value of urea and creatinine level was statistically significant higher among patients with renal failure and inflammation than controls. The mean value of urea and creatinine level was statistically significant higher among patients with renal failure and inflammation than patients with renal failure. The mean value of urea and creatinine level was statistically significant higher among patients with renal failure than controls. There were statistically significant positive correlations between IL-18 and (Glucose and CRP), while negative correlations between IL-18 and albumin. **Conclusion:** IL-18 levels were elevated in renal failure patients than controls. Serum IL-18 is significantly increase in renal failure patients and is correlated with inflammation. So, IL-18 can be used as non-invasive pro inflammatory marker for detection of the chronicity and severity in renal failure patients.

KEYWORDS: Il-18- renal failure-end stage renal disease.

INTRODUCTION

End-stage renal disease (ESRD) last period of chronic kidney disease led to a substantial rise in cardiovascular disease and infection morbidity and mortality, reflecting respective 20 and 50 percent of gross death rates; ESRD patients.[1] The risk for infection is high for patients who suffer from hemodialysis (HD). This is because inadequate cell immunity increases their infectious resistance.[2] Interleukin-18 (IL-18), a powerful multi-property proinflammatory cytokine, is a member of the cytokine IL-1 family, which was primarily referred to as the inductive factor of interferons gamma produced in several types of cells alternately .It participates in cellular and humeral reactions.[3]

In addition to its pro-inflammatory role, IL-18 participates by its direct impact on the renal function of nephropathy.[4]

The aim of this study was to evaluate the role of pro-inflammatroy cytokine interleukin-18 in end stage renal disease.

MATERIAL AND METHODS

This prospective a case-control study which conducted on 150 subjects all collected from Benha teaching Hospital and were divided into three groups Group (1): control group composed of 50 healthy volunteer individuals. Group (2): 50 patients with renal failure. Group (3): 50 patients with renal failure and inflammation. This study was conducted during the period from November 2018 to November 2020. Samples of studied groups were analyzed at Molecular Biology Unit of Medical Biochemistry Department Benha teaching Hospital.

All subjects were subjected to the following: Full history, Clinical examination, Laboratory investigations:

The sample was divided into two parts

Part 1: put on EDTA for complete blood count (CBC) including the differential a count was done automatically (Symex, XS- 800i japan)

Part 2: was configured for minutes at 1000xg hyperlipidemic and hemolzed sample were excluded – serum were used for the following biochemical investigations tests

1. Kidney profile test (Creatinine – Albumin – uric acid – urea – Na^+ and K^+)
2. Glucose and CRP
3. Human Interleukin 18 (IL-18). ELISA Kit Human Interleukin 18 (IL-18). ELISA Kit Catalog Number. CSB-E07450h. For the quantitative determination of human interleukin 18 (IL-18) concentrations.

Ethical considerations

The committee of ethics of scientific research of Menofia Faculty of science approved the studied protocol and written consents were obtained from studied groups for participation in this study.

PRINCIPLE OF THE ASSAY

This assay employs the quantitative sandwich enzyme immunoassay technique. Antibody specific for IL-18 has been pre-coated onto a microplate. Standards and samples are pipetted into the wells and any IL-18 present is bound by the immobilized antibody. After removing any unbound substances, a biotin-conjugated antibody specific for IL-18 is added to the wells. After washing, avidin conjugated Horseradish Peroxidase (HRP) is added to the wells. Following a wash to remove any unbound avidin-enzyme reagent, a substrate solution is added to the wells and color develops in proportion to the amount of IL-18 bound in the initial step. The color development is stopped and the intensity of the color is measured.

Statistical analysis

The data were coded, entered and processed on computer using *Statistical package for social science (SPSS)* (version 24).The results were represented in tabular and diagrammatic forms then interpreted. Mean, standard deviation, range, frequency, and percentage were use as descriptive statistics. The following test was done: Chi-Square test X^2 was used to test the association variables for categorical data. Student's t-test was used to assess the statistical significance of the difference between two population means in a study involving independent samples. ANOVA (F test) for normally quantitative variables, to compare between more than two groups, and Post Hoc test (LSD) for pairwise comparisons. Pearson's correlation coefficient: it evaluates the linear association between 2 quantitative variables (one is the independent var. X, and the other is the dependent var., Y). Value of "r" ranges from -1 to 1. P value was considered significant as the following: $P > 0.05$: Non significant, $P \leq 0.05$: Significant

RESULTS

This table showed that, the mean of age in controls, patients with renal failure, patients with renal failure and inflammation were 48.27 ± 4.49, 49.10 ±5.04 and 49.90 ±1.6 years respectively. There was no statistically significant difference between studied groups regarding age **(Table 1)**.

This study showed that, the percentage of males among controls was 60%, while 64% among patients with renal failure and 62% among patients with renal failure and inflammation. There is amale predominance. There was no statistically significant difference between studied groups regarding sex **(Table 2)**.

This table showed that, the mean of IL-18 in controls, patients with renal failure, patients with renal failure and inflammation were 98.50±60.01, 548.78±140.73 and 1114.16±243.03 respectively. The mean value of IL-18 was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001.The mean value of IL-18 was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001.The mean value of IL-18 was statistically significant higher among patients with renal failure than controls and P value <0.001 **(Table 3)**.

This table showed that, that the mean of creatinine level in controls, patients with renal failure, patients with renal failure and inflammation were 0.93±0.27, 11.56±8.85 and 12.59±4.24 respectively. The mean value of creatinine level was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001. The mean value of creatinine level was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001. The mean value of creatinine level was statistically significant higher among patients with renal failure than controls and P value <0.001 **(Table 4)**.

This table showed that, that the mean of urea level in controls, patients with renal failure, patients with renal failure and inflammation were 23.98±6.29, 92.86±30.06 and 132.92±14.42 respectively. The mean value of urea level was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001. The mean value of urea level was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001. The mean value of urea level was statistically significant higher among patients with renal failure than controls and P value <0.001 **(Table 5)**.

This table showed that the correlation among IL-18 and other variables in CRP-negative group. There were statistically significant positive correlations between IL18 and (Urea mg/dl, Uric acid mg/dl, Glucose mg/dl).

There were no statistically significant correlations between IL18 and other numerical data **(Table 6)**.

This table showed that the correlations among IL-18 and other variables in CRP –positive group. There were statistically significant positive correlations between IL18 and (Creatinine mg/dl, Glucose mg/dl, CRP). There were no statistically significant correlations between IL18 and other numerical data **(Table 7)**.

Table (1): Age among different groups

		Controls (n.= 50)	CRP negative (n.= 50)	CRP –positive (n.= 50)	F	p
Age (years)	**Rang**	20 - 70	20 - 75	20 - 75	-0.402	0.689
	Mean ± SD	48.27 ± 4.49	49.10 ±5.04	49.90 ±1.6		

Table (2): Sex among different groups

			Controls (n.= 50)	CRP negative (n.= 50)	CRP –positive (n.= 50)	X^2	p
Sex	**female**	**No.**	20	18	19	0.067	0.795
		%	40%	36%	38%		
	male	**No.**	30	32	31		
		%	60%	64%	62%		

Table (3): Level of IL-18 among different groups.

Variable	Controls (n.= 50)	CRP negative (n.= 50)	CRP –positive (n.= 50)	F	p
	Mean ± S.D (Std. Deviation) (minimum – maximum)				
IL-18 (Pg/ml) (Min – Max)	98.50±60.01 (20.44 – 194.00)	548.78±140.73 (309.45 – 798.63)	1114.16±243.03 (111.48 – 1917.70)	471.063	<0.001

Table (4): Creatinine level among different groups.

Variable	Controls (n.= 50)	CRP negative (n.= 50)	CRP –positive (n.= 50)	F	p
	Mean ± S.D (Std. Deviation) (minimum – maximum)				
Creatinine (mg/dl) (Min – Max)	0.93±0.27 (0.60 – 1.40)	11.56±8.85 (6.10 – 16.8)	12.59±4.24 (6.10 – 19.50)	64.88	<0.001

Table (5): Urea level among different groups

Variable	Controls (n.= 50)	CRP negative (n.= 50)	CRP –positive (n.= 50)	F	p
	Mean ± S.D (Std. Deviation) (minimum – maximum)				
Urea (mg/dl) (Min – Max)	23.98±6.29 (15.00 – 45.00)	92.86±30.06 (27.00 – 143.00)	132.92±14.42 (58.00 – 149)	20.34	<0.001

Table (6): Correlations among IL-18 and other variables in CRP-negative group.

Variable	IL18	P value
Hb (g/dl)	-0.058	0.58
Creatinine (mg/dl)	0.166	0.16
Urea (mg/dl)	0.811	<0.001
Uric acid (mg/dl)	0.642	<0.001
ALB (gm/dl)	-0.097	0.50
Glucose (mg/dl)	0.631	<0.001
Sodium (mmol/l)	-0.170	0.23
Potassium (mmol/l)	0.046	0.75

Table (7): correlations among IL-18 and other variables in CRP –positive group.

	IL-18	P-value
Hb (g/dl)	-0.141	0.32
Creatinine (mg/dl)	0.550	<0.001
Urea (mg/dl)	0.093	0.52
Uric acid (mg/dl)	0.172	0.23
ALB (gm/dl)	-0.007	0.96
Glucose (mg/dl)	0.335	0.018
Sodium (mmol/l)	-0.150	0.29
Potassium (mmol/l)	0.136	0.34
CRP	0.430	0.002

DISCUSSION

This study showed that, the mean of age in controls, patients with renal failure, patients with renal failure and inflammation were 48.27 ± 4.49, 49.10 ±5.04 and 49.90 ±1.6 years respectively. There was no statistically significant difference between studied groups regarding age and sex.

This study showed that the mean of IL-18 in controls, patients with renal failure, patients with renal failure and inflammation were 98.50±60.01, 548.78±140.73 and 1114.16±243.03 respectively. The mean value of IL-18 was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001.The mean value of IL-18 was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001.The mean value of IL-18 was statistically significant higher among patients with renal failure than controls and P value <0.001.

This finding was in accordance with the study of **Makled et al.,**[1] who found that, there is high elevation of IL-18 in ESRD than controls.

In harmony with the present study, **Yong et al.,**[5] who found that, IL18 was elevated in CKD in comparison to healthy controls.

Fujita et al.,[4] in Japan and **Liu et al.,**[6] in China demonstrated that there was an elevation of IL-18 mRNA and protein in urine and serum of patients with ESRD with infection, which supports the use of cytokines as markers that mirror the progression of microinflammation and incidence of ESRD.

This result was in agreement with **Miyamoto et al.,**[7] who found serum levels of IL-18 levels are often several times higher in ESRD patients than in healthy controls.

This study showed that, that the mean of creatinine level in controls, patients with renal failure, patients with renal failure and inflammation were 0.93±0.27, 11.56±8.85 and 12.59±4.24 respectively. The mean value of creatinine level was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001.The mean value of creatinine level was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001.The mean value of creatinine level was statistically significant higher among patients with renal failure than controls and P value <0.001.This study showed that, that the mean of urea level in controls, patients with renal failure, patients with renal failure and inflammation were 23.98±6.29, 92.86±30.06 and 132.92±146.42 respectively. The mean value of urea level was statistically significant higher among patients with renal failure and inflammation than controls and P value <0.001.The mean value of urea level was statistically significant higher among patients with renal failure and inflammation than patients with renal failure and P value <0.001.The mean value of urea level was statistically significant higher among patients with renal failure than controls and P value <0.001.

This is in agreement with **Mansour et al.,**[8] who aimed to assess the relationship between serum Mg levels and vascular calcification in chronic hemodialysis (HD) patients. A cross-sectional study conducted on 60 patients with end-stage renal disease on regular HD in Al-Zahraa University Hospital (group I) compared with 30 healthy controls (group II), from June to December 2017. They found that, there were highly significant increase regarding blood urea, serum creatinine in group I compared with group II.

This agrees also with **Hsu et al.,**[9] who found that, negative impact of infection events during post-ESRD outcome expand on the emerging theory that pre-ESRD conditions, such as abrupt decline in kidney function (creatinine and urea level), have a sustained impact on post-ESRD outcome.

This study showed that, there was no statistically significant correlation between IL-18 and hemoglobin among the studied groups.

In contrast to these results, **Makled et al.,**[1] who found, there was a positive correlation between Hb and IL-18 serum levels.

In the current study, there were statistically significant positive correlations between IL-18 and Glucose.

This agrees with **Makled et al.,**[1] who found, there was a positive correlation between serum glucose and IL-18 serum levels.

This was in accordance with that reported by **Esposito et al.,**[10] who detected that hyperglycemia produces an acute rise of IL-18 concentrations.

This study showed that, there were statistically significant positive correlations between IL-18 and CRP. In harmony with the present study, **Liu et al.,**[11] who conducted a cohort study to examine the IL-18 level among hemodialysis patients. They found that, IL-18 levels, as a continuous variable, showed modest correlations with CRP levels, suggesting that IL-18 may play a role in the ESRD patients.In a study of 102 ESRD patients on maintenance dialysis, **Porazko et al.,**[12] demonstrated a strong association between IL-18 and CRP $r = 0.24$; $p < 0.05$).

In our study, there were statistically significant negative correlations between IL-18 and albumin.

This agrees with **Formanowicz et al.,**[13] who found that there is significant reverse relationship between IL-18 and albumins.

This result was comparable to result documented by **Marcelli et al.,**[14] who found patients with relatively high serum IL-18 had lower protein intake. Moreover, IL-18 was found to be negatively correlated with albumin.

CONCLUSION

IL-18 levels were elevated in renal failure patients than controls. Serum IL-18 is significantly increase in renal failure patients and is correlated with inflammation. So, IL-18 can be used as non-invasive pro inflammatory marker for detection of the chronicity and severity in renal failure patients. Thus, the present study attempts to gain a better understanding of the involvement of IL-18 in renal failure.

REFERENCES

1. Makled AFM, Salem EHA, El-Khayat AHA, El Shohady SIM. Interleukin-18 and vascular endothelial growth factor as predictors for end-stage renal disease. Menoufia Med J, 2017; 30(8): 1014-21.
2. Bernieh B. Viral hepatitis in hemodialysis: an update. J Transl Int Med, 2015; 3(3): 93-105.
3. Bhat OM, Dhawan V. Role of IL-18 and its signaling in atherosclerosis. Inflamm Cell Sig, 2015; 12(3): 7-17.
4. Fujita T, Ogihara N, Kamura Y, Satomura A, Fuke Y, Shimizu C, Wada Y, Matsumoto K. Interleukin-18 contributes more closely to the progression of diabetic nephropathy than other diabetic complications. Acta diabetologica, 2012; 49(2): 111–17.
5. Yong K, Ooi EM, Dogra G, Mannion M, Boudville N., Chan D, Lim E M, Lim W H. Elevated interleukin-12 and interleukin-18 in chronic kidney disease are not associated with arterial stiffness. Cytokine, 2013; 64(1): 39–42.
6. Liu F, Guo J, Zhang Q, Liu D, Wen L, Yang Y, Yang L, Liu Z. The Expression of Tristetraprolin and Its Relationship with Urinary Proteins in Patients with Diabetic Nephropathy. PloS one, 2015; 10(10): 0141471.
7. Miyamoto T. Carrero JJ; Stenvinkel P. Inflammation as a risk factor and target for therapy in chronic kidney disease. Curr Opin Nephrol Hypertens, 2011; 20(6): 662–668.
8. Mansour HH, Mohamed RR, Khaled MF, Khalil EMSerum magnesium level in maintenance hemodialysis and cardiovascular calcification. Sci J Al-Azhar Med Fac Girls, 2018; 2(3): 26-31.
9. Hsu R K, Chai B, Roy J A, Anderson A H, Bansal N, Feldman HI, Go AS, He J, Horwitz E J, Kusek JW, Lash J P, Ojo A, Sondheimer J H, Townsend RR, Zhan M, Hsu CY. CRIC Study Investigators. Abrupt Decline in Kidney Function before Initiating Hemodialysis and All-Cause Mortality: The Chronic Renal Insufficiency Cohort (CRIC) Study. American journal of kidney diseases: the official journal of the National Kidney Foundation, 2016; 68(2): 193–202.
10. Esposito K, Nappo F, Marfella R, Giugliano G, Giugliano F, Ciotola M, Quagliaro L, Ceriello A, Giugliano D. Inflammatory cytokine concentrations are acutely increased by hyperglycemia in humans: role of oxidative stress. Circulation, 2002; 106(16): 2067–72.
11. Liu Y-W, Su C-T, Chang Y-T, Tsai W-C, Su Y-R, Wang SPH Elevated Serum Interleukin-18 Level Is Associated with All-Cause Mortality in Stable Hemodialysis Patients Independently of Cardiac Dysfunction. PLoS ONE, 2014; 9(3): 89-94.
12. Porazko T, Kúzniar J, Kusztal M, Kúzniar T J, Weyde W, Kuriata-Kordek M, Klinger M. IL-18 is involved in vascular injury in end-stage renal disease patients. Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association, 2009; 24(2): 589–96.
13. Formanowicz D., Wanic-Kossowska M., Pawliczak E., Radom M., Formanowicz P., Usefulness of serum interleukin-18 in predicting cardiovascular mortality in patients with chronic kidney disease systems and clinical approach. Sci Rep, 2015; 5(1): 18-32.
14. Marcelli D, Wabel P, Wieskotten S, Ciotola A, Grassmann A, Di Benedetto A, Canaud B. Physical methods for evaluating the nutrition status of hemodialysis patients. J Nephrol, 2015; 28(5): 523–30.