Int. j. adv. multidisc. res. stud. 2023; 3(3):1092-1095

Received: 07-05-2023
Accepted: 17-06-2023

International Journal of Advanced Multidisciplinary Research and Studies

ISSN: 2583-049X

IL-12 Pathway as Prognostic Factor in Lupus Nephritis Disease

[1] Doaa K Hussein, [2] Mayyada F Darweesh

[1, 2] Department of Microbiology, Faculty of Science, University of Kufa, Iraq

Corresponding Author: **Mayyada F Darweesh**

Abstract

Background: Lupus nephritis (LN) is a major complication of SLE affect 50-60% of SLE patients that affects around 5 million people worldwide. IL-12/pathway is a pivotal player in the immune system and is central to controlling number of inflammatory diseases. This study design to investigate the role of IL-12/ as biomarker in lupus nephritis patients.

Materials and Methods: A case-control study design on 70 participates which included 30 lupus nephritis patients with different stages diagnosis by nephrology specialist collected from nephrology department in Al-sadder medical city in AL- Najaf, from October 2022 till end of February 2023 and 40 control groups divided into two types, first 20 apparently healthy and 20 nephropathy patients without autoimmunity. Blood sample was collected from all participants to detected IL-12/ serum level by enzyme linked immunosorbent assay (ELISA).

Results: The results showed a significant increase in the mean levels of serum IL-12 in lupus nephritis patients (37.09 ± 3.47) were higher than healthy and nephropathy patients (1.89 ± 0.47; 12.32± 3.41 pg/ml) respectively, the difference was highly significant (P <.0001). According to lupus nephritis grades, IL-12 serum level increase with disease progression, so IL-12 in grade IV higher than grade I**.**

Conclusion: Serum levels of IL-12/were significantly increased in lupus nephritis Iraqi patients and increased correlated with progressive of disease may be considered as a prognostic factor for disease.

Keywords: Lupus Nephritis, IL-12

Introduction

Lupus nephritis (LN) Is a severe form of SLE characterized by subendothelial and/or subepithelial immune complex depositions in the affected kidney, resulting in significant injury and nephron loss during the acute phase and subsequently chronic irreversible damage and renal function impairment if not treated successfully [1]. Lupus development is aided by deficiencies in innate and adaptive immunity, Autoantibodies directed against nuclear and cellular antigens are often generated, resulting in immune complex development and immune complex accumulation in glomeruli [2]. These autoantibodies have the following features in relation to lupus nephritis: Anti-dsDNA antibodies may react with the basement membrane of the glomerulus, Autoantibodies with a higher affinity may form intravascular immune complexes that deposit in the glomeruli, Cationic autoantibodies bind to the anionic basement membrane more strongly and Autoantibodies of certain isotypes activate complements [3]. Cytokine-mediated inflammatory processes have been implicated in the development of acute kidney injury (AKI) and chronic kidney disease (CKD), where endothelium and tissue damage are connected with the release of particular mediators that may launch the inflammatory cascade. Cytokines may have a predictive role in addition to their role in disease etiology [4]. The interleukin 12 (IL-12) family consists of four members IL-12, IL-23, IL-27, and IL-35. IL-12 is a pro-inflammatory cytokine serve as a link between the innate and adaptive immune systems which induces Th cells differentiation into Th1 cells through induction of IFN γ that is essential for the development of different autoimmune diseases, such as experimental autoimmune uveitis [5, 6]. An increase in IL-12 concentration has been observed in patients with SLE compared with healthy controls, and this is positively associated with the SLE disease activity index (SLEDAI) [7].

Materials and Methods

Patients and Control characterization

A case-control study design on 70 participates which included 30 patients with lupus nephritis, the age of patients range (11-70) with different stages of LN collected from nephrology department in Al-sader medical city in AL-Najaf, from October 2022 till end of February 2023 and 40 control groups divided into two types, first 20 apparently healthy with the age range(11-70) and 20 nephropathy patients with the age rang (13-70)years.

Sample Collection

Four ml was collected from both patients and controls was placed into a gel tube. After the blood had been at room temperature for about 30 minutes to allow for clotting, it was centrifuged for 10 minutes at 5000 rpm, then serum separated and stored at 20 C until used for measuring IL-12/ according to (Elabscience, USA).

Statistical Analysis

Statistical analysis was carried out by using statistical software (IBM SPSS Statistics 26)**.** The result of IL-12 level were expressed as arithmetic mean ± SE. The comparison between patients and control groups was analyzed by one-way ANOVA (more than two means). P-value (<0.05) was considered significant statistically.

Ethical Committee

Ethical approval: The study was conducted in accordance with ethical principles that have their origin in the Declaration of Helsinki. It was performed with patients' consent both verbally and analytically before sampling. The study protocol, subject information, and consent form were reviewed and approved by the local ethics committee according to the document number 397 in 19/1/2023 to get this approval.

Results

Distribution of Patients and Control Subjects According to Type of Disease, Age and Gender

A case-control results appear that group of lupus nephritis patients included more females 22 than male 8**,** 20 apparently healthy with male: female 10: 10 for each one and 20 nephropathy patients as shown in table (1).

According to means age of patients lupus nephritis group recoded 36.03 ±2.19 years while healthy subjects recorded 33.7 ±3.14 years versus 31.6 ±3.15 for nephropathy patients group.

Table 1: Distribution of study subjects according to type of disease, age, gender and Grades

	Autoimmune based glomerulonephritis	**control group**	
	Lupus nephritis N= 30	Healthy group N= 20	Nephropathy group N=20
Male	8(26.6%)	10 (50%)	12 (60%)
Female	22(73.3%)	10 (50%)	8 (40%)
Age mean±SE	36.03 ±2.19	33.7 ±3.14	31.6 ±3.15
Age range	11-70	11 -70	13-70
< 20	2 (6.66%)	4 (20%)	3 (15%)
20-44	12 (40%)	8 (40%)	6 (30%)
>45	16 (53.3%)	6 (30%)	8 (40%)
Lupus nephritis Grades	Grade I	6 (20%)	
	Grade III	11 (36.6%)	
	Grade IV	13(43.3%)	

Evaluation Serum IL-12 Levels in Patients with Lupus Nephritis and Control

The results showed that serum IL-12 levels were significantly higher in lupus nephritis patients (37.09 ± 3.47 pg/ml) than in the control groups (12.32± 3.41pg/ml; 1.89 ± 0.89 pg/ml) respectively at (P= 0.0001) As show in(Table 2).Also the mean of IL-12 serum level was greater in the progression of disease, grade IV (48.47 ± 3.86 pg/ml) higher as compared to grade I and grade III lupus nephritis (19.13 ± 6.93 pg/ml; 33.45 ± 5.21) respectively at (P 0.003)**.** As show in (Table 2) and figure (1):

Table 2: Mean of IL-12 serum level in lupus nephritis patients compared control groups

Study group	**IL-12**	**P-value**
Lupus nephritis (30)	37.09 ± 3.47	0.0001***
Nephropathygroups (20)	12.32± 3.41	
Healthy groups (20)	1.89 ± 0.47	
Grade I	33.45 ± 5.21	0.003**
Grade III	19.13 ± 6.93	
Grade VI	48.47 ± 3.86	

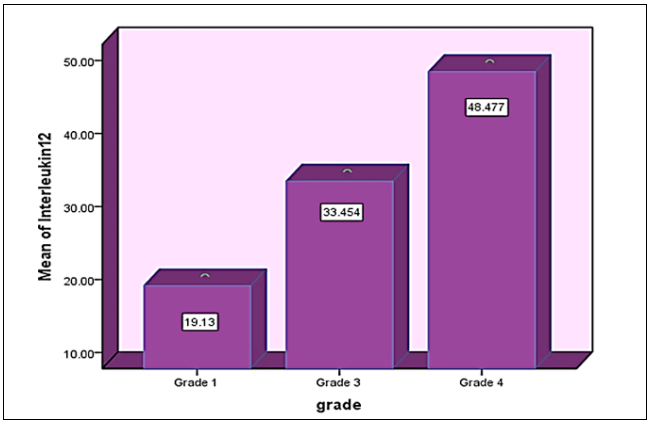

Fig 1: IL-12 serum level in lupus nephritis patients according to grade

Discussion

This study found that females outnumbered males in LN patients, which is similar with a local study in Baghdad [11], which found that females outnumbered males, with females 32(80%) and males 8(20%) with ages ranging from 19 to 44 years. Also in Thi-Qar -Iraq research [12] discovered that SLE is caused by chronic and recurring activation of the immune system, affecting more females (46%) than males (8%), with a mean of age (range) 22.1(12-40) years. Another study [13] proposed that the incidence of SLE in females was 100%, which could be due to the potential roles of genetic variations on the X chromosome and sex hormone milieus, as well as sex differences in environmental exposures or sensitivity to these exposures, as potential risk factors of incident SLE. According to the nephropathy group this result confirmed with [14] they found that male sex was a risk factor for the development of nephropathy and that women had higher nephron protection than males when other risk variables such as hypertension and albuminuria were controlled too. Also, [15] discovered that nephropathy progressed in men than in females, and the possible mechanisms behind the renal protective function of the female gender appear to be connected to estrogen hormone. Notably, in other studies, women had a greater prevalence of nephropathy than males. This might be due to the fact that most women were postmenopausal, therefore these findings could be impacted by the loss of estrogen-mediated nephron-protection. The current study showed that serum IL-12 level were significantly higher in lupus nephritis patients than in the control groups. Also, the mean of IL-12/ serum level was greater in the progression of disease. This similar to [16], they found SLE patients significantly higher levels of IL-12, compared to healthy control

(HC)($p < 0.05$).another study [17] they Found the plasma level of IL-12 in SLE patients was significantly greater than in healthy controls (19.64± 5.65 vs 13.2 ±7 6.27 (pg/ml), P 0.001). Indicate that increased IL-12 production is associated closely with renal disease in parallel with Th1 polarization. In line with [18] they found that active SLE patients, particularly those with nephritis, display an increased serum IL and the concentration in the blood correlated with disease activity. dysregulation of the IL-12 cytokine contributes to inhibition of Treg activity and promotion of MHC expression, Th17 differentiation, T/B-cell activation and survival, and autoantibody production [19]. Also In Iraq research [20] they observed Significantly higher levels of IL-12 in the SLE group ($p < 0.001$) compared to normal control(Median IL-12 (pg/ml) 309.92 - 165.75) respectively, Because IL-12 activates the JAK/STAT pathway, it promotes the differentiation of naïve CD4+ T cells into IFN- y producing Th1 cells as well as the differentiation of T follicular helper cells (Tfh). This result confirmed with [21] they observed level of plasma cytokine (IL-12) in SLE patients compared to the healthy controls are highly significantly. As a result, our findings add to previously research showing that renal flares are associated with an IFN-γ signature. Overexpression of IL-12 causes further inflammation and tissue injury and contributes to the immunopathogeneses of SLE[23].This result is agreement with [24] They divided Patients into two groups: inactive patients (class I, II, and III) and active patients (class IV and V). IL-12 levels were lower in patients with inactive SLE than in patients with active SLE, suggesting that IL-12 reflected the illness severity of SLE patients. Also [26] they observed the level of IL-12 difference according to classes of lupus nephritis patients (3.4±0.67, 4± 0.74, and 5.6 ± 0.43) in classes(II, IV, and V respectively).

References

1. Parodis I, Tamirou F, Houssiau FA. Prediction of prognosis and renal outcome in lupus nephritis. Lupus Sci. Med. 2020; 7(1):pe000389. Doi: 10.1136/lupus-2020-000389
2. Parikh SV, Almaani S, Brodsky S, Rovin BH. Update on lupus nephritis: Core curriculum 2020, Am. J. Kidney Dis. 2020; 76(2):265-281.
3. Musa R, Brent LH, Qurie A. Lupus Nephritis. StatPearls Publishing, Treasure Island (FL), 2022. [Online]. Available: http://europepmc.org/books/NBK499817
4. Ortega LM, Fornoni A. Role of cytokines in the pathogenesis of acute and chronic kidney disease, glomerulonephritis, and end-stage kidney disease, Int. J. Interf. Cytokine Mediat. Res. 2010; 2:49-62.
5. Sun L, He C, Nair L, Yeung J, Egwuagu CE. Interleukin 12 (IL-12) family cytokines: Role in immune pathogenesis and treatment of CNS autoimmune disease, Cytokine. 2015; 75(2):249-255.
6. Yang P, Foster CS. Interleukin 21, interleukin 23, and transforming growth factor β1 in HLA-A29-associated birdshot retinochoroidopathy, Am. J. Ophthalmol. 2013; 156(2):400-406.
7. Huang X, Hua J, Shen N, Chen S. Dysregulated expression of interleukin-23 and interleukin-12 subunits in systemic lupus erythematosus patients, Mod. Rheumatol. 2007; 17(3):220-223.
8. Castro F, Cardoso AP, Gonçalves RM, Serre K, Oliveira MJ. Interferon-gamma at the crossroads of tumor immune surveillance or evasion, Front. Immunol. 2018; 9:p847.
9. Pollard KM, Cauvi DM, Toomey CB, Morris KV, Kono DH. Interferon-γ and systemic autoimmunity, Discov. Med. 2013; 16(87):p123.
10. Chalmers SA, Garcia SJ, Reynolds JA, Herlitz L, Putterman C. NF-kB signaling in myeloid cells mediates the pathogenesis of immune-mediated nephritis, J. Autoimmun. 2019; 98:33-43.
11. Ibrahim N, Allawi A, Ghudhaib K, Hammoudi FA. Estimation of some immunological markers of Iraqi patients in systemic lupus erythematosus with lupus nephritis, Prof. (Dr) RK Sharma. 2020; 20(4):p4668.
12. Sabr NH, Obaid KA, Nisan AH. Hepatomegaly, Splenomegaly and lymphadenopathy in Iraqi patients with Systemic Lupus Erythematosus, Univ. Thi-Qar J. Med. 2020; 19(1):1-11.
13. Woo JMP, Parks CG, Jacobsen S, Costenbader KH, Bernatsky S. The role of environmental exposures and gene-environment interactions in the etiology of systemic lupus erythematous, J. Intern. Med. 2022; 291(6):755-778.
14. Skupien J, *et al*. Variations in risk of end-stage renal disease and risk of mortality in an international study of patients with type 1 diabetes and advanced nephropathy, Diabetes Care. 2019; 42(1):93-101.
15. Riispere Ž, *et al*. IgA nephropathy clinicopathologic study following the Oxford classification: Progression peculiarities and gender-related differences, Medicina (B. Aires). 2016; 52(6):340-348.
16. Salazar-Camarena DC, *et al*. BAFF-R and TACI expression on CD3+ T cells: Interplay among BAFF, APRIL and T helper cytokines profile in systemic lupus erythematosus, Cytokine. 2019; 114:115-127.
17. Ye QL, Guoliu RN, Qin H, Shen YY, Wang B, Zhai ZM. Elevated plasma levels of IL-12 and IFN-γ in systemic lupus erythematosus, Int J Clin Exp Pathol. 2017; 10(3):3286-3291.
18. Ma X, *et al*. Expansion of T follicular helper-T helper 1 like cells through epigenetic regulation by signal transducer and activator of transcription factors, Ann. Rheum. Dis. 2018; 77(9):1354-1361.
19. Mok CC. Targeted Small Molecules for Systemic Lupus Erythematosus: Drugs in the Pipeline, Drugs, 2023, 1-18.
20. Saud BARAM. Role of Interleukin-12 in Pathogenesis of Systemic Lupus Erythematosus, Iraqi J. Biotechnol. 2022; 21(2).
21. Uzrail AH, Assaf AM, Abdalla SS. Correlations of expression levels of a panel of genes (IRF5, STAT4, TNFSF4, MECP2, and TLR7) and Cytokine Levels (IL-2, IL-6, IL-10, IL-12, IFN-γ, and TNF-α) with systemic lupus erythematosus outcomes in Jordanian patients, Biomed Res. Int, 2019.
22. Munroe ME, *et al*. Altered type II interferon precedes autoantibody accrual and elevated type I interferon activity prior to systemic lupus erythematosus classification, Ann. Rheum. Dis. 2016; 75(11):2014-2021.
23. Liu W, Li M, Wang Z, Wang J. IFN-γ mediates the development of systemic lupus erythematosus, Biomed

Res. Int, 2020.
24. Li L, Sun X, Wu S, Yuan X, Liu B, Zhou X. Interleukin-12 exacerbates symptoms in an MRL/MpJ-Faslpr mouse model of systemic lupus erythematosus, Exp. Ther. Med. 2021; 21(6):1-8.
25. Elewa EA, Zakaria O, Mohamed EI, Boghdadi G. The role of interleukins 4, 17 and interferon gamma as biomarkers in patients with Systemic Lupus Erythematosus and their correlation with disease activity, Egypt. Rheumatol. 2014; 36(1):21-27.
26. Hassan LA, Majeed AA, Darweesh MF. The Role of IL-12 in the Aetiology of SLE and its Connection to HBV Infection in Iraqi Patients, J. Commun. Dis. (E-ISSN 2581-351X P-ISSN 0019-5138). 2022; 54(1):41-46.