Role of Interleukin-17A and Interleukin-23 as Biomarkers in Rheumatoid Arthritis Patients

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ABSTRACT

Background: Cytokines are crucial in the mechanisms underlying inflammation and articular degeneration accompanying the autoimmune disease rheumatoid arthritis. Interleukine-17 and interleukine-23 were shown to have a key function in the chronic immune process and the consequent tissue destruction in the joints of rheumatoid patients.

Aims: The study goal was to quantify interleukins-17A and IL-23 concentrations in a group of newly diagnosed rheumatoid patients and to evaluate if there is a linkage between these cytokines and the activity parameters of this illness.

Subjects and Methods: A study involving 250 individuals, 125 rheumatoid patients, and 125 controls was performed. The ELISA technique utilized Blood samples to estimate interleukines-17A and -23.

Results: Rheumatoid cases had higher IL-17A (median= 49.7 pg/ml) and IL-23 (mean= 21.1 ng/L) concentration, $P < 0.05$, than controls. IL-17A was shown to be positively correlated with DAS-28 ($P<0.01$), C-reactive protein ($r= 0.036$, $P < 0.01$), and ESR value ($r = 0.021$, $P < 0.05$). The AUC for IL-17A= 0.742, and IL-23=0.804 with $P < 0.05$).

Conclusion: Both interleukine17-A and IL-23 possess value in the pathogenesis of rheumatoid arthritis, and it is possible to use them as biomarkers for disease diagnosis and to predict the prognosis of the disease.

Keywords: Rheumatoid arthritis, cytokine, interleukins, biomarker.

دور الانترلوكين ١٧-أ والانترلوكين ٢٣ كمؤشرات حيوية في مرضى التهاب المفاصل الرثوي

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الخلاصة

الخلفية: تعتبر السايتوكينات لها دور اساسي في عملية الالتهاب والضمور العضلي المصاحب لمرض المناعة الذاتية والتهاب المفاصل الرثوي. تبين أن إنترلوكين١٧-أ وإنترلوكين -٢٣ لهما وظيفة رئيسية في عملية المناعة المزمنة بالإضافة إلى تدمير الأنسجة الناتجة في المفاصل المصابة لدى مرضى التهاب المفاصل الرثوي.

الأهداف: كان هدف الدراسة هو قياس تركيزات الإنترلوكينات ١٧-أ و إنترلوكين -٢٣ في مجموعة من مرضى التهاب المفاصل الرثوي الذين تم تشخيصهم حديثاً وأيضاً لتقييم ما إذا كان هناك ارتباط بين هذه السيتوكينات ومعلمات النشاط لهذا المرض.

المواد والطرق: أجريت دراسة شملت ٢٥٠ فرداً، ١٢٥ مريض التهاب المفاصل الرثوي و١٢٥ مجموعة قياسية. تم استخدام عينات الدم لتقدير الإنترلوكينات١٧-أ و-٢٣، عبر تقنية ايلايزا.

النتائج: كان لحالات التهاب المفاصل الرثوي تركيزات أعلى من انترلوكين ١٧-أ (الوسيط= ٤٩.٧ بيكوغرام/مل) و إنترلوكين-٢٣ (المتوسط= ٢١.١ نانوغرام/لتر)، ($P <0.05$)، مقارنةً بالضوابط. ارتبط انترلوكين ١٧-أ بشكل إيجابي مع DAS-28 ($P <0.01$) ، والبروتين التفاعلي C، $P <0.01$ ($r = 0.036$) وقيمة ESR، $P <0.05$ ($r = 0.021$). و كانت المساحة تحت المنحني للانترلوكين ١٧-أ =٠.٧٤٢، و للانترلوكين ٢٣=٠.٨٠٤ ، ($P <0.05$).

الاستنتاج: نستنتج ان كلا من الإنترلوكين ١٧-أ والإنترلوكين -٢٣ لهما قيمة في التسبب في التهاب المفاصل الرثوي وهناك إمكانية لاستخدامهما كمؤشرات حيوية لتشخيص المرض والتنبؤ بعواقبه.

الكلمات المفتاحية: التهاب المفاصل الرثوي، السايتوكين، الإنترلوكينات، العلامات الحيوية.

INTRODUCTION

Rheumatoid arthritis (RA) is an illness that leads to permanent joint degeneration, severe disability, and early death[1]. This autoimmune disease is characterized by attracting neutrophils and T cells to the inflammation area. T helper-17 (Th-17) lymphocytes, a particular subgroup of CD4+ T lymphocytes, release characteristic cytokines, including IL-17, -21, and -22, and they were shown to have a key function in the persistent inflammatory process in rheumatoid patients joints[2].

Interleukine-23 (IL-23), a pro-inflammatory mediator belonging to the IL-12 cytokine family, is produced via several activated immune system cells as monocytes, dendritic cells, and macrophages. For Th-17 lymphocytes to differentiate from naïve CD4+ T lymphocytes, IL-23 is necessary[3]. Th-17 cells generate IL-17 and other mediators when influenced by IL-23, and both IL-17 and IL-23 have been linked to the production of autoimmune tissue inflammation together with autoimmune conditions like rheumatoid arthritis[4]. Through two separate pathways, IL-23 is crucial in persistent inflammation, a characteristic feature of various autoimmune disorders. The first method involves Th-17 cell activation, and the second involves non-T cells inducing IL-17 production[5].

Th-17 lymphocytes, after stimulation by IL-23, will secrete IL-17 cytokine that is a robust osteoclastogenesis promoter, primarily through initiation of activation of nuclear factor-κB ligand (RANKL) receptor, the fundamental enhancer for osteoclastocyte expansion that intensifies the corrosion in the border of the joints in individuals with severe rheumatoid disease[6]. IL-17 stimulates the synthesis of inflammatory mediators like tumor necrosis factor-α (TNF-α) and IL-1, found in the synovial fluid and serum of RA patients. IL-17A is an effective producer of several pro-inflammatory cytokines; it has the potential to cause joint inflammation as well as bone and cartilage loss; it boosts IL-6 production which causes collagen breakdown and reduces synovium and cartilage collagen synthesis as well as cartilage proteoglycan synthesis[7]; Thus, IL-23 has a significant effect in determining the synthesis of IL-17A, a cytokine that has an impact on inflammation and bone loss in RA [8].

IL-17A block in RA mouse models reduced disease progression, leading to diminished joint irritation and bone corruption, indicating that it is an essential mediator of RA pathogenesis. As a result, anti-IL-17A cytokine therapy is being investigated as a potential anti-rheumatic strategy for RA [9].

The Objectives of the current research are to investigate the serum concentrations of both IL-17A and IL-23 in rheumatoid cases and controls, explore relationships of these cytokines to several disease parameters, and determine the possibility of utilizing them as biomarkers in the identification of the disease.

MATERIALS AND METHODS

Ethical Approval

Ethical approval was obtained from Nineveh Health Directorate (Number: 46690 on 29/12/2021 -the research protocol number is 198/21). The College of Medicine at the University of Mosul's "Medical Research Ethics Committee (MREC)" gave its approval for this study on December 27, 2021 (Ref. no: UOM/COM/MREC/ 21-22(21). Before joining the study, each subject gave written consent.

Subjects

From December 2021 to September 2023, A case-control study was performed in which 125 RA cases attending an outpatient rheumatic clinic at Al-Salam Teaching Hospital, Mosul City, Nineveh Province, Iraq, were included as the test group. 125 healthy (thoroughly assessed by the clinician as healthy subjects) were included as controls. Match in terms of age and gender were chosen. Patients enrolled in the work had been diagnosed with rheumatoid clinically by a specialized rheumatologist. RA is categorized using the modified 1987 ACR/ EULAR guidelines[10]. This research excluded other inflammatory joint disorders, autoimmune diseases, and vasculitis cases. One commonly used metric to evaluate disease activity in RA is the 28-joint score (DAS28). The twenty-eight (wrists, knees, feet, and proximal interphalangeal and metacarpophalangeal) joints were examined for swelling and tenderness as part of a specialist doctor's clinical evaluation.

Blood sampling and quantification of serum IL-17A and IL-23 level

Each subject provides three ml of whole blood in a gel tube to coagulate. After centrifugation, the serum was isolated and saved at -20°C till the analysis. As directed by the manufacturer's guidelines, IL-17A and IL-23 concentrations were tested utilizing a commercial-specific ELISA kit (Shanghai YL Biont, China, Catalog No: YLA1538HU, Human IL-17 ELISA kit, Shanghai YL Biontt, China, Catalog No: YLA0681HU, Human IL-23 ELISA ki). Briefly, 40µl of serum samples were added to the microplate ELISA wells, and then 50µl streptavidin-HRP was added. Then, the plate was kept in an incubator at 37°C (60 min). Following washing, each hole received 50 µl of colored complex solution A, followed by 50 µl of colored complex solution B; after that, the holes were incubated for about ten min at 37°C. Each hole received 50 µl of stopping solution to bring the reaction to an end. Finally, every well's wavelength was calculated at 450 nm immediately using an ELISA reader. From a calibration graph that mapped the absorption value against quantity, optical density was obtained from each reference standard in pg/ml for IL-17A (R_2 = 0.993) and ng/L for IL-23(R_2 = 0.94).

Statistical Analysis

Categorical information was presented using frequency and percentages. Continuous data, on the other hand, were represented by the mean ± standard deviation (Mean ± SD). The Kolmogorov-Smirnov assessment is employed to calculate the normality test of continuous data. A non-parametric Mann-Whitney measure was applied to measure statistical validity. Spearman's Rho linear association analysis was utilized to analyze the statistical validity of linear correlation between two numerical variables. The serum IL-17A and IL-23 function was assessed utilizing receiver-operating characteristics (ROC) graphs. Statistical examination in this research was carried out using MedCalc® Statistical Tools (version 19.1). The degree of significance (P value) was < 0.05.

RESULTS

This study involves 250 subjects divided into two sets; the first included 125 RA cases with ages ranging from (21- 65) years and a Mean ± SD of (42.8±10.9) years. Their illness duration was (10.7±1.8) years. The second group is the control group, which included 125 healthy individuals whose ages range from (22-67) years and a Mean ± SD (45.6±8.6) years. Table 1 summarizes the demographic details of the RA cases participating in this work.

Our results reveal that RA cases had meaningfully greater both IL-17A and IL-23 concentrations compared to controls (median = 49.7 pg./ml against 29.3 pg./ml for IL-17A, and mean = 21.1 ng/L for IL-23, P < 0.05), (figure 1).

A high statistically substantial difference between IL-17A concentration regarding DAS-28 activity score in addition to a highly statistically significant variation in IL-17A median between RA individuals who have an excellent disease activity score and patients with RA that are in remission (P<0.01) was found, whereas no statistically significant change was found regarding IL-23 cytokine and RA disease activity scores (P>0.05), as demonstrated in figure 2.

Table 1: Data and disease characteristics of Rheumatoid Arthritis Subjects

Parameter	Number	%
Age (Mean ± SD) = (42.8±10.9) Years		
Age group		
<30 years	46	36.8%
31-49 years	48	38.4%
≥ 50 years	31	24.8%
RA disease duration (years) ± SD = (10.7±1.8) Years		
DAS-28 Mean ± SD = (6.22±1.32)		
High disease activity	42	33.6%
Modest disease activity	34	27.2%
Low disease activity	32	25.6%
Remission	17	13.6%
ESR of RA patients Mean ± SD = 42±22.59 mm/hr.		
CRP of RA patients Mean ± SD = 12.32±10.72 mg/L		
Rheumatoid factor (RF)	Positive: 85(68%)	Negative: 40(32%)
Total	125	100.0%

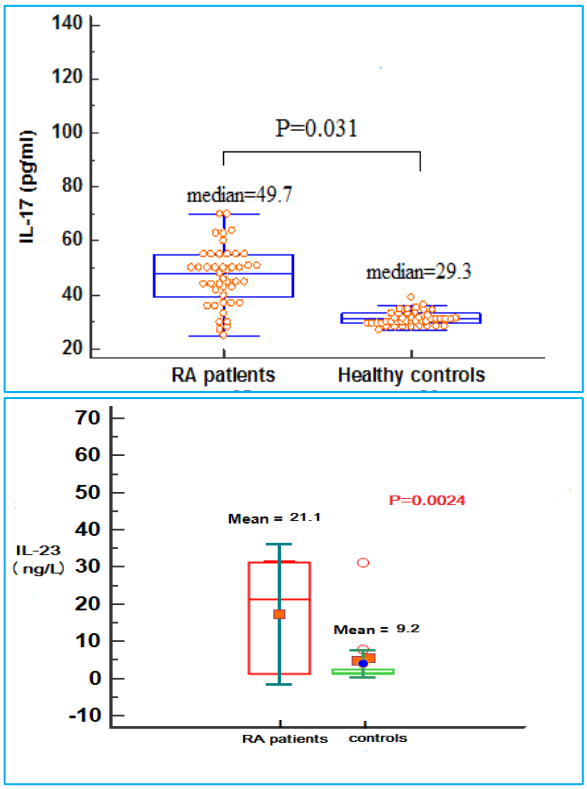

Figure 1: IL-17A and IL-23 quantities in rheumatoid versus control subjects (P<0.05).

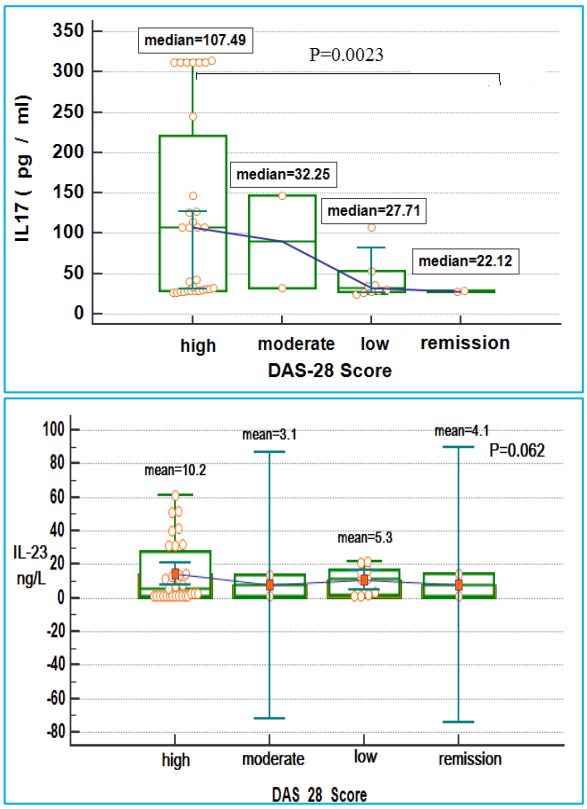

Figure 2: DAS-28 scores and IL-17A and IL-23 levels in rheumatoid cases (P<0.01).

A positive association was observed between the values of IL-17A and specific inflammatory illness indicators as ESR (r - 0.021, P < 0.05), positive RF (P < 0.05), and C-reactive protein (r - 0.036, P < 0.01), as shown in figure 3. In comparison, no statistically significant association between IL-23 and illness indicators (P > 0.05) was found (results not shown).

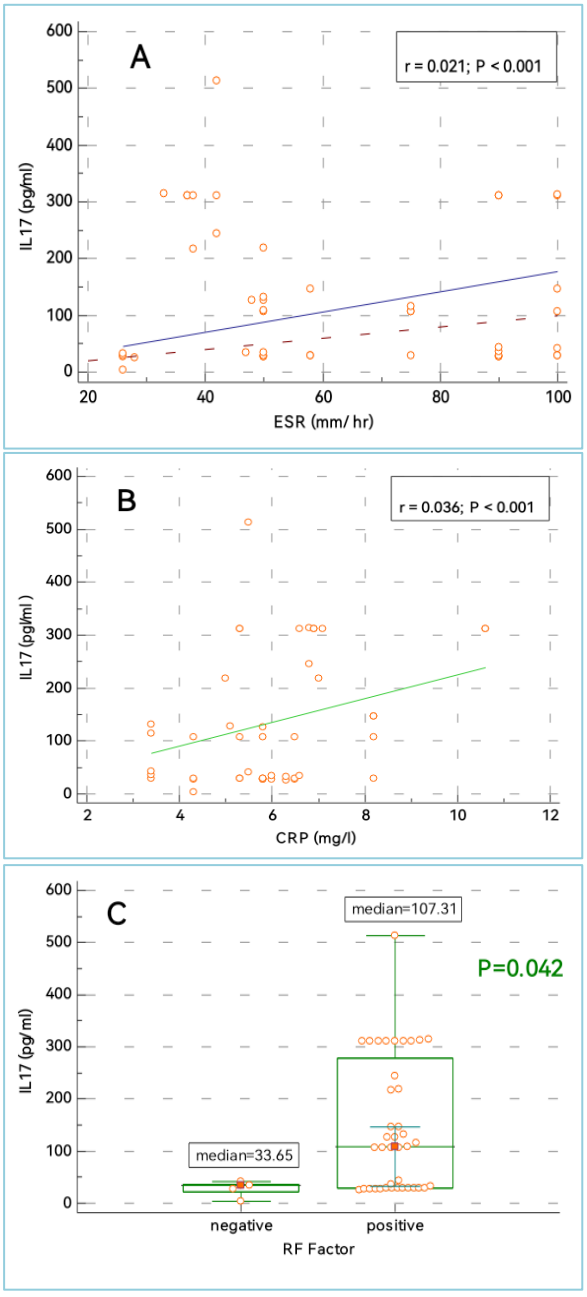

Figure 3: Serum IL-17A value expresses a positive relationship with several inflammatory indicators: A: ESR (r - 0.021, P < 0.05), B: C-reactive protein (r - 0.036, P <0.01), C: positive RF (P < 0.05).

To examine the capability of the studied cytokines in RA diagnosis, a ROC graph was used; the part below the curve (AUC) for IL-17A at value <39.2 pg/ml (Optimum cut-off) was 0.742 and exhibited 48.9% sensitivity and 97.8% specificity while the AUC for IL-23 at value 27.7 was 0.804 and has 92.5 % sensitivity and 20% specificity (P < 0.05). as demonstrated in figure 4.

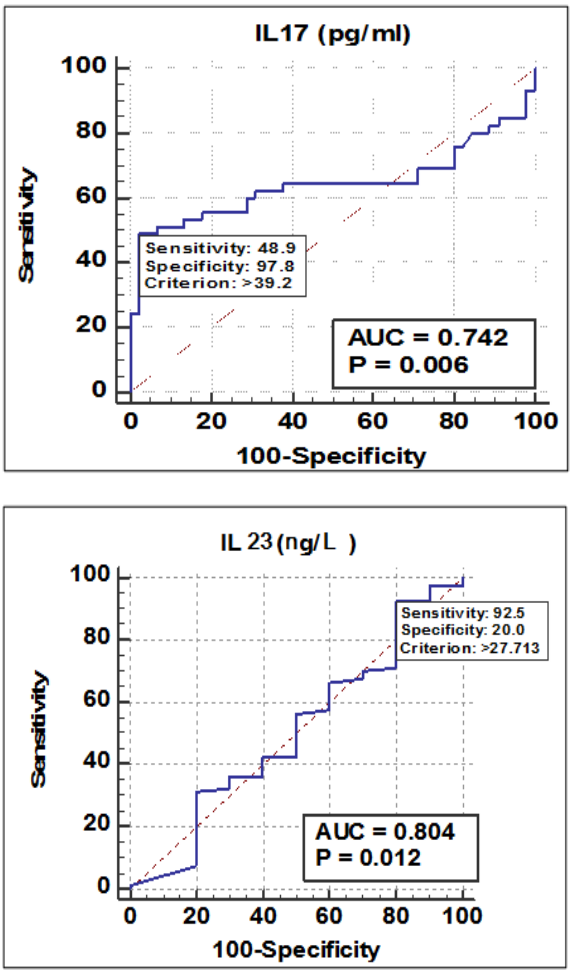

Figure 4: IL-17A and IL-23 ROC graph for evaluating Rheumatoid arthritis.

DISCUSSION

Rheumatoid arthritis patients are frequently misdiagnosed due to the varied clinical presentations and absence of characteristic manifestations, in addition to negative serological tests in the initial disease stages[11].

Recent research shows how crucial IL-17 and Th-17 lymphocytes are to initiating RA. IL-17 cytokine is a newly discovered pro-inflammatory protein involved in joint damage and RA inflammatory process. The Th-17 axis has emerged as a particular target in the therapy of RA, and numerous treatments that target Th-17 cells or their effector mediators are being investigated[12].

IL-17A also facilitates the manufacture of prostaglandin E2, TNF-α, IL-1, -6, and -8, which aid in neutrophil attraction to the site of inflammation[12].

Findings revealed a substantial increase in IL-17A in the serum of RA patients compared to healthy people, which goes with other studies[13,14]. It was recently discovered that Th-17 cell activation and high IL-17 production are typical features of RA [12]. There was a substantial variance between the studied subgroups concerning DAS-28 score and IL-17A levels, which were their highest levels among RA patients who had severe activity scores than those in remission; this goes with the results of Kim et al. and Atwa et al.[15,16]. Results of the above research found that IL-17 and Th-17 cells are essential in RA etiology, and the elevated concentrations of IL-17A in the circulation are linked to higher activity scores and articular degenerative symptoms in RA patients. According to earlier animal model studies, IL-17A increases disease activity in arthritis patients by acting independently of TNF-α [17].

Research accomplished by Farag et al. and Al-Saadany et al.[18,19] discovered a substantial association between parameters of disease severity "CRP, ESR, RF, DAS-28" and the mean serum concentration of IL-17, which coincide with present study findings, this indicates the relevance of IL-17A in characterizing disease severity. Additionally, IL-17 is a potent C-reactive protein stimulator, suggesting that serum IL-17A had a significant function in the damaging pattern that characterizes RA. IL-17 induces cartilage damage by increasing synoviocyte migration, chemokine expression, invasiveness, and metalloproteinase secretion; it also facilitates angiogenesis[20].

The pro-inflammatory cytokine IL-23 is essential for Th-17 cell growth. Furthermore, it has been shown that the IL-17/IL-23 axis is crucial in persistent destructive arthritis[21]. This study shows a significant elevation in IL-23 concentration in the serum of rheumatoid patients versus controls (P<0.05), a result which goes with previous studies as a study conducted in Egypt by Zaki and colleagues[22] and several other studies[23-27] that show increased IL-23 concentration in RA patients. Thus, our findings support the general agreement that IL-23 may be a valuable biomarker for diagnosing RA.

Findings reveal no connection between IL-23 and disease activity scores despite the slight increase in IL-23 concentrations in high disease activity patients; however, this increase is not statistically significant (P>0.05), a result that goes inside with other studies[8]. However, other studies have found inconsistent results[24,26]. Variations in the RA patients enrolled in the different studies and the joint X-ray data may explain these conflicting findings. Additionally, no correlation was found between IL-23 and other disease parameters such as ESR, RF factor, and CRP. It has been found that following anti-TNF-α medication, responders had a substantial drop in IL-23 concomitant with clinical remission, indicating the value of this mediator in rheumatoid pathogenesis[28].

CONCLUSIONS

Results imply both IL-17A and IL-23 cytokines were crucial in pathogenesis of RA and possess a diagnostic utility in this illness. In patients with RA, elevated IL-17A quantities in the blood coincide simultaneously with the severity and illness activity. This may demonstrate IL-17A's relevance as a potential biomarker for joint injury, providing it a significant predictive utility. The knowledge that IL-17A and IL-23 were crucial cytokines engaged in RA disease pathogenicity makes them an attractive prospective treatment candidate for RA. Future pharmaceutical treatments for RA focusing on the IL-17A beside IL-23 axis have been investigated and show promise.

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Conflict of Interest

The authors declare no competing interests.

REFERENCES

1. Bullock J, Rizvi SAA, Saleh AM, Ahmed SS, Do DP, Ansari RA, et al. Rheumatoid Arthritis: A Brief Overview of the Treatment. Medical principles and practice : international journal of the Kuwait University, Health Science Centre. 2018;27(6):501-7. DOI: 10.1159/000493390
2. Hull DN, Cooksley H, Chokshi S, Williams RO, Abraham S, Taylor PC. Increase in circulating Th17 cells during anti-TNF therapy is associated with ultrasonographic improvement of synovitis in rheumatoid arthritis. Arthritis Res Ther. 2016;18(1):303.DOI:10.1186/s13075-016-1197-5
3. Sisto M, Lisi S. Interleukin-23 Involved in Fibrotic Autoimmune Diseases: New Discoveries. Journal of clinical medicine. 2023;12(17). doi: 10.3390/jcm12175699
4. Bunte K, Beikler T. Th17 Cells and the IL-23/IL-17 Axis in the Pathogenesis of Periodontitis and Immune-Mediated Inflammatory Diseases. Int J Mol Sci. 2019;20(14). DOI: 10.3390/ijms20143394
5. Izcue A, Hue S, Buonocore S, Arancibia-Cárcamo CV, Ahern PP, Iwakura Y, et al. Interleukin-23 Restrains Regulatory T Cell Activity to Drive T Cell-Dependent Colitis. Immunity. 2008;28(4):559-70. DOI: 10.1016/j.immuni.2008.02.019
6. Lin D, Li L, Sun Y, Wang W, Wang X, Ye Y, et al. IL-17 regulates the expressions of RANKL and OPG in human periodontal ligament cells via TRAF6/TBK1-JNK/NF-κB pathways. Immunology. 2014;144(3):472-85. doi: 10.1111/imm.12395
7. Chen J, Liao MY, Gao XL, Zhong Q, Tang TT, Yu X, et al. IL-17A induces pro-inflammatory cytokines production in macrophages via MAPKinases, NF-κB and AP-1. Cellular physiology and biochemistry: international journal of experimental cellular physiology, biochemistry, and pharmacology. 2013;32(5):1265-74. DOI: 10.1159/000354525
8. Zaky DS, El-Nahrery EM. Role of interleukin-23 as a biomarker in rheumatoid arthritis patients and its correlation with disease activity. International immunopharmacology. 2016;31:105-8. DOI: 10.1016/j.intimp.2015.12.011
9. Taams LS. Interleukin-17 in rheumatoid arthritis: Trials and tribulations. The Journal of Experimental Medicine. 2020;217(3). DOI: 10.1084/jem.20192048
10. Arnett FC, Edworthy SM, Bloch DA, McShane DJ, Fries JF, Cooper NS, et al. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. Arthritis and rheumatism. 1988;31(3):315-24. DOI: 10.1002/art.1780310302

11. Heidari B. Rheumatoid Arthritis: Early diagnosis and treatment outcomes. Caspian Journal of Internal Medicine. 2011;2(1):161-70.
12. Wang J, He L, Li W, Lv S. A Role of IL-17 in Rheumatoid Arthritis Patients Complicated With Atherosclerosis. Frontiers in Pharmacology. 2022;13. https://doi.org/10.3389/fphar.2022.828933
13. Hitchon CA, Alex P, Erdile LB, Frank MB, Dozmorov I, Tang Y, et al. A distinct multi cytokine profile is associated with anti-cyclical citrullinated peptide antibodies in patients with early untreated inflammatory arthritis. J Rheumatol. 2004;31(12):2336. https://www.jrheum.org/content/jrheum/31/12/2336.full.pdf.
14. Metawi SA, Abbas D, Kamal MM, Ibrahim MK. Serum and synovial fluid levels of interleukin-17 in correlation with disease activity in patients with RA. Clinical rheumatology. 2011;30(9):1201-7. DOI: 10.1007/s10067-011-1737-y
15. Kim J, Kang S, Kim J, Kwon G, Koo S. Elevated levels of T helper 17 cells are associated with disease activity in patients with rheumatoid arthritis. Annals of laboratory medicine. 2013;33(1):52-9. DOI: 10.3343/alm.2013.33.1.52
16. Atwa SE, Azab MM, Mohamed MS, El Sheikh mM. Serum Interleukin-17 Level in Patients with Rheumatoid Arthritis and its Relation to Disease Activity. Zagazig University Medical Journal. 2020;26(1):87-93. DOI: 10.21608/zumj.2019.11577.1196
17. Adamopoulos IE, Chao CC, Geissler R, Laface D, Blumenschein W, Iwakura Y, et al. Interleukin-17A upregulates receptor activator of NF-kappaB on osteoclast precursors. Arthritis Res Ther. 2010;12(1):R29. DOI: 10.1186/ar2936
18. Farag MA, El Debaky FE, Abd El-Rahman SM, Abd el-khalek SM, Fawzy RM. Serum and synovial fluid interleukin-17 concentrations in rheumatoid arthritis patients: Relation to disease activity, radiographic severity and power Doppler ultrasound. The Egyptian Rheumatologist. 2020;42(3):171-5. DOI: 10.1016/j.ejr.2020.02.009
19. Al-Saadany HM, Hussein MS, Gaber RA, Zaytoun HA. Th-17 cells and serum IL-17 in rheumatoid arthritis patients: Correlation with disease activity and severity. The Egyptian Rheumatologist. 2016;38(1):1-7. https://doi.org/10.1016/j.ejr.2015.01.001
20. Koenders MI, Marijnissen RJ, Devesa I, Lubberts E, Joosten LA, Roth J, et al. Tumor necrosis factor-interleukin-17 interplay induces S100A8, interleukin-1β, and matrix metalloproteinases, and drives irreversible cartilage destruction in murine arthritis: rationale for combination treatment during arthritis. Arthritis and rheumatism. 2011;63(8):2329-39. DOI: 10.1002/art.30418
21. Iwakura Y, Ishigame H. The IL-23/IL-17 axis in inflammation. The Journal of Clinical Investigation. 2006;116(5):1218-22. DOI:10.1172/JCI28508
22. Zaky DSE, El-Nahrery EMA. Role of interleukin-23 as a biomarker in rheumatoid arthritis patients and its correlation with disease activity. International immunopharmacology. 2016;31:105-8. DOI: 10.1016/j.intimp.2015.12.011
23. Dalila AS, Mohd Said MS, Shaharir SS, Asrul AW, Low SF, Shamsul AS, et al. Interleukin-23 and its correlation with disease activity, joint damage, and functional disability in rheumatoid arthritis. The Kaohsiung Journal of Medical Sciences. 2014;30(7):337-42. DOI: 10.1016/j.kjms.2014.02.010
24. Guo YY, Wang NZ, Zhao S, Hou LX, Xu YB, Zhang N. Increased interleukin-23 is associated with increased disease activity in patients with rheumatoid arthritis. Chinese medical Journal. 2013;126(5):850-4. DOI: 10.3899/jrheum.100259
25. Wendling D, Abbas W, Godfrin-Valnet M, Kumar A, Guillot X, Khan KA, et al. Dysregulated Serum IL-23 and SIRT1 Activity in Peripheral Blood Mononuclear Cells of Patients with Rheumatoid Arthritis. PloS one. 2015;10(3):e0119981. https://doi.org/10.1371/journal.pone.0119981
26. Alsheikh MM, El-Shafey AM, Gawish HH, El-Desoky ET. Serum interleukin-23 level in rheumatoid arthritis patients: Relation to disease activity and severity. The Egyptian Rheumatologist. 2019;41(2):99-103. https://doi.org/10.1016/j.ejr.2018.07.001
27. Abu Al Fadl EM, Fattouh M, Allam AA. High IL-23 level is a marker of disease activity in rheumatoid arthritis. The Egyptian Journal of Immunology. 2013;20(2):85-92. https://ejimmunology.org/pdf/2013-june/8-Mona%20Fatouh.pdf
28. Kageyama Y, Ichikawa T, Nagafusa T, Torikai E, Shimazu M, Nagano A. Etanercept reduces the serum levels of interleukin-23 and macrophage inflammatory protein-3 alpha in patients with rheumatoid arthritis. Rheumatology international. 2007;28(2):137-43. DOI: 10.1007/s00296-007-0388-4