

## Diagnostic values of serum IL-17A and IL-18 in *Rheumatoid arthritis* and Correlation with treatment

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### Abstract

**Background:** Rheumatoid arthritis (RA) is an autoimmune disorder that involves autoantibodies attacking and weakening joints. RA is characterized by leukocyte (Monocyte, Lymphocyte mast cell .etc) infiltrations into the synovial compartment leading to inflammation in the synovial membrane. Synovitis leads to the release of pro-inflammatory cytokines, matrix metalloproteinases, chemokines, complement proteins, and growth factors.

**Objective:** The current study pointed to verify the diagnostic values of interleukin -17 A and interleukin -18 in *Rheumatoid arthritis* (RA) patients and the effect of treatment thereon.

**Study subjects and methods:** A total of 88 samples with RA were selected from the health clinics of AL-Yarmouk teaching hospital/rheumatology clinic in Baghdad, with female rheumatoid arthritis patients as the patient group (50) and (38) healthy females as the control group. All patients were exposed to clinical, laboratory, and ultrasound assessments, besides measuring the serum level of both (IL-17A and IL-18) by the method of Enzyme-linked immunosorbent assay (ELISA).

**Results:** The results show that there is a significant difference ( $p \leq 0.05$ ) in IL-17A levels between patients and controls. The concentration of IL-17A in premenopausal patients is higher when compared to control groups ( $18.06 \pm 3.85$  vs  $15.71 \pm 1.82$  pg/ml), so, the concentration in postmenopausal studied groups ( $17.19 \pm 2.91$  vs  $14.13 \pm 1.06$  pg/ml).

Also, there are significant differences ( $p \leq 0.05$ ) in the level of IL-18 between the patients and the control; it is found that the level of IL-18 within the premenopausal patients was higher compared to the control ( $16.09 \pm 9.69$  vs  $12.52 \pm 8.30$  pg/ml).

**Conclusion:** A High level of IL-17A in RA patients contributes to the pathogenesis of RA as an inflammatory disease. As well as, the elevated levels of IL-18 suggest its physiological role to induce inflammatory disorder Treatment with (MTX and Etanercept) causes a decrease in the inflammatory markers of this disease RF in patient groups.

**Keywords:** Rheumatoid, arthritis, Interleukin -17A, Interleukin -18, correlation, treatment

### Introduction

*Rheumatoid arthritis* is an autoimmune condition, which means that the individual's immune system attacks itself in an uncontrolled way, this attack is directed against the joints, and contributes to their gradual degradation, resulting in chronic inflammation (1). Progressive disease stages can lead to extensive loss of mobility and functioning; it starts with inflammation of joint areas in the body, particularly in the small bones of the feet and fingers (2). If there are an excessively large number of inflammatory cells, damages body tissue. Female

reproductive factors, including menopause and hormonal therapy, show an influential role in the severity of the disease. On the other hand, it was found that pregnancy is associated with reduced illness activity in half of the women due to immunosuppression during this time, but the postpartum period is associated with. Innate immune cells such as mast cells and neutrophils participate in the development, and so do macrophages that work by releasing pro-inflammatory interleukins (such as TNF- alpha, IL-1, and IL-6) (3). Cartilage destruction is partly encouraged by synovial cytokines like IL-17

Investigates on RA synovial fluids display that IL-17 stimulates the secretion of IL6, Leukemia inhibitory factors, (LIF), and macrophages inflammatory proteins (MIP) (4). In response to TNF stimulation, RA synovial tissue fibroblast releases biological active IL-18. Interestingly, in reply to IL-18 activation, which consists of adhesion molecules, neutrophil chemo-attractant, and monocyte chemo-

#### Material and methods

This study included (88) individuals, (50) of the study sampling were patients, and (38) individuals were studied as healthy control. The samples were collected from at AL-Yarmouk teaching hospital/Rheumatology clinic.

Five mL of fasting venous blood was obtained from the study subjects, agglutinated at room temperature for about 20 minutes. The serum was obtained by centrifugation at 3000 rpm for 10 minutes.

#### Results and discussion

##### The level of IL-17A in the studied groups:

The current study publicized that there were significant differences ( $p \leq 0.05$ ) in the IL-17A level between patients and controls. The concentration of

attractant, several factors are secreted from RA fibroblasts, suggesting that IL-18 contributes to the pathogenesis of RA through various approaches (5).

**Aim of the study:** Determining the level of some immunological cytokines (IL-17A, IL-18) in RA female patients and studying the effect of immunological and biochemical parameters on the disease activity.

All patients were exposed to clinical, laboratory biochemical parameters such as ESR erythrocyte sedimentation rate and immunological as Anti-cyclic citrullinated peptide and Rheumatoid factor, and ultrasound assessment and to the measurement of IL-17 and IL-18 in the serum by ELISA method following the manufacturer instructions (IL-17 kit, Diaclone / France; IL-18 kit, Al-shkairate / Gordon).

IL-17A in premenopausal patients was higher compared to premenopausal control ( $18.06 \pm 3.85$  vs  $15.71 \pm 1.82$  pg/ml), as so as, it is concentration in postmenopausal studied groups was ( $17.19 \pm 2.91$  vs  $14.13 \pm 1.06$ ) as illustrated in figure (1).

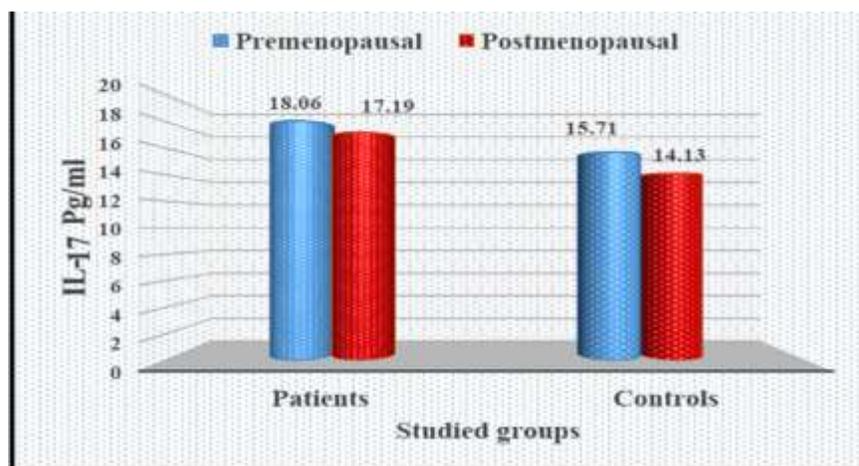


Figure (1): The level of IL-17 in the studied groups, Showed the premenopausal and postmenopausal effects

##### The level of IL-18 in the studied groups

In the current study, there was a significant difference ( $p \leq 0.05$ ) in IL-18 levels between patients and the control, the level of IL-18 in premenopausal patients compared to the control ( $16.09 \pm 9.69$  vs

$12.52 \pm 8.30$  pg/ml), also, in the postmenopausal studied group the level of IL-18 was higher in postmenopausal female patients compared to the control group ( $20.30 \pm 14.8$  vs  $11.70 \pm 5.77$  pg/ml) as illustrated in Figure (2).

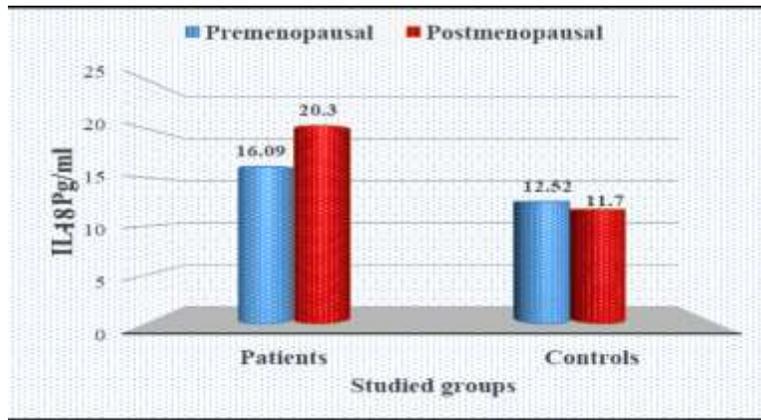


Figure (2): The level of IL-18 in the studied groups, Showed that premenopausal And postmenopausal effects

**Effect of treatment on the concentration of RF and Anti-CCP in patients:**

In this study, there was a significant difference ( $p \leq 0.05$ ) in the level of RF between the patients who has received methotrexate (MTX) and biotherapy

such as Etanercept (Enbrel) with patients who did not receive therapy for 6 months, the concentration of RF in patients received therapies were lower compared with patients who did not ( $26.0 \pm 23.3$  vs  $131 \pm 25.8$  U/ml) as shown in Figure (3).

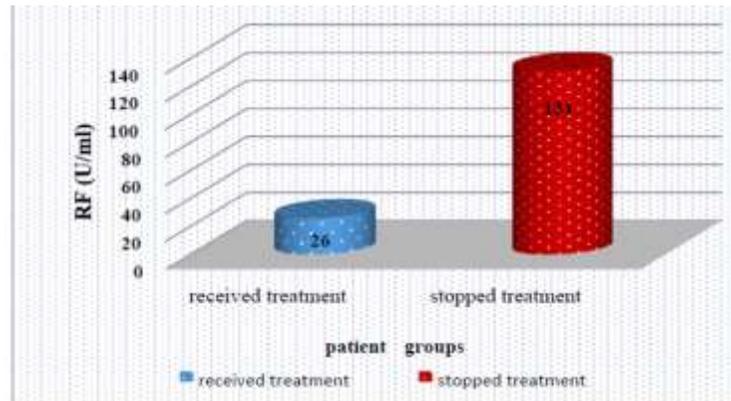
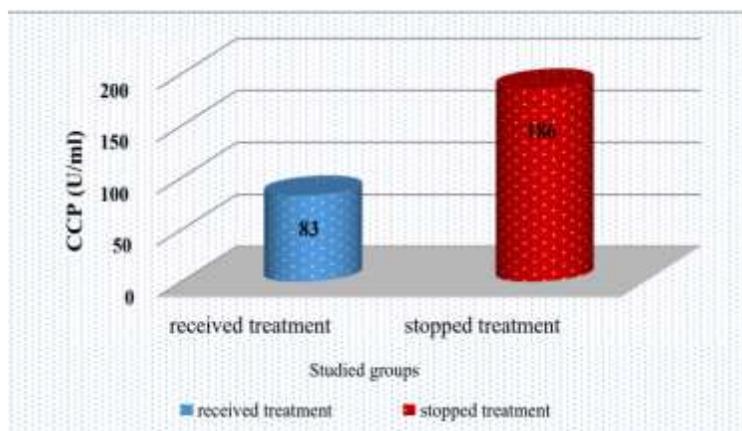


Figure (3): Effect of MTX and biotherapy on the RF concentration

In this result the concentration of anti-CCP in the female patients who received MTX and ETN (Enbrel) was lower than its concentration in female

patients were did not ( $83 \pm 14.1$  vs  $186 \pm 17.3$  U/ml) as shown in figure (4), there were significant differences ( $p \leq 0.05$ ).



**Figure (4): Effect of MTX and biotherapy on the anti-CCP concentration**

### Discussion

The results in figure (1), were agreed with (6) who observed that the synovial IL-17A serum level was found at high titer in RA patients with active disease, and extra-articular appearances like dry eyes, dry mouth, and subcutaneous nodules. Other reports by

In this study, there were significant differences ( $p \leq 0.05$ ) in the concentration of RF between the patients who has received methotrexate (MTX) and biotherapy such as Etanercept (Enbrel) with patients who did not receive therapy for 6 months, the concentration of RF in patients received therapies were lower compared with patients who did not ( $26.0 \pm 23.3$  vs  $131 \pm 25.8$  U/ml) as shown in figure (3).

A severe decrease in the level of CCP was observed in patients with rheumatoid arthritis who were treated

### Conclusion

A high level of IL-17A in RA patients promotes the pathogenesis of RA as an inflammatory disease. As well as, the elevated levels of IL-18 suggest its physiological role to induce inflammatory disorder.

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(7) revealed that IL-17 expression was significantly higher in RA synovium compared with the control.

The results in figure (2), were agreed with (8) who showed that the IL-18 serum level was constitutively high in RA patients when compared with the controls.

with TNF inhibitors, including ETN drug, accompanied by a similar improvement in disease activity as the disease activity that obtained 28 points (DAS28) (4).

Results indicated that usage with both ETN and MTX drugs offered more returns in terms of the clinical indications and signs, inflammatory keys, and autoantibody titers (9).

Treatment with (MTX and Etanercept) causes a decrease in the inflammatory markers of this disease in RF and anti-ccp autoantibodies in the patients' groups.

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## القيم التشخيصية لمصل IL17A و IL18 في التهاب المفاصل الروماتويدي وعلاقته بالعلاج

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### الخلاصة

خلفية البحث: التهاب المفاصل الروماتويدي (RA) هو اضطراب مناعي ذاتي يشمل الأجسام المضادة الذاتية التي تهاجم المفاصل وتضعفها. يتميز التهاب المفاصل الروماتويدي بارتشاح الكريات البيض (الخلية الوحيدة ، الخلايا البدينة للمفاوية .. إلخ) إلى الحيز الزليلي مما يؤدي إلى التهاب الغشاء الزليلي. يؤدي التهاب الغشاء المفصلي إلى إطلاق السيتوكينات المحفزة للالتهابات ، والبروتينات المعدنية المصفوفة ، والكيموكينات ، والبروتينات التكميلية ، وعوامل النمو.

الهدف: أشارت الدراسة الحالية إلى التحقق من القيم التشخيصية للإنترلوكين -17 ألف والإنترلوكين -18 في مرضى التهاب المفاصل الروماتويدي وتأثير العلاج فيها.

مواضيع الدراسة وطرقها: تم اختيار مجموعه مؤلفة من 88 عينة مصابة بالتهاب المفاصل الروماتويدي من العيادات الصحية في مستشفى اليرموك التعليمي / عيادة أمراض الروماتيزم في بغداد ، شملت الدراسة (50) من مرضى التهاب المفاصل الروماتويدي الاناث و (38) من الإناث الأصحاء كمجموعة ضابطة. تم إخضاع جميع المرضى للتقييم السريري والمختبري والموجات فوق الصوتية ، إلى جانب قياس مستوى المصل لكل من-IL (IL-17A و IL-18) بطريقة الممتز المناعي المرتبط بالإنزيم (ELISA) .

النتائج: أظهرت النتائج أن هناك فرقا معنويا ( $p \leq 0.05$ ) في مستوى IL-17A بين المرضى والمجموعة الضابطة. يكون تركيز IL-17A في مرضى ما قبل انقطاع الطمث أعلى عند مقارنته بالمجموعة الضابطة ( $3.859 \pm 18.06$  مقابل  $1.82 \pm 15.71$  بيكوغرام / مل) ، وتركيزه في مرضى بعد انقطاع الطمث ( $2.91 \pm 17.19$  مقابل  $1.06 \pm 14.13$  بيكوغرام/ مل).

أيضا ، هناك اختلافات معنوية كبيرة ( $p \leq 0.05$ ) في مستوى IL-18 بين مجموعة المرضى و المجموعة الضابطة ؛ فقد وجد أن مستوى IL-18 لدى مرضى ما قبل انقطاع الطمث كان أعلى مقارنة بالمجموعة الضابطة ( $9.69 \pm 16.09$  مقابل  $8.30 \pm 12.52$  بيكوغرام / مل). الخلاصة: ارتفاع مستوى IL-17A في مرضى التهاب المفاصل الروماتويدي يساهم في التسبب في التهاب المفاصل الروماتويدي باعتباره مرضاً التهابياً. بالإضافة إلى ذلك ، تشير المستويات المرتفعة من IL-18 إلى دوره الفسيولوجي في إحداث الاضطراب الالتهابي. يؤدي العلاج باستخدام (MTX و Etanercept) إلى انخفاض في العلامات الالتهابية لهذا المرض RF في مجموعات المرضى.

الكلمات المفتاحية : الروماتويدي، التهاب المفاصل، الإنترلوكين -17 ، الإنترلوكين-18 ، الارتباط، العلاج.