Estimation of IL-17 levels in sera of Iraqi patients with TB and COVID-19 infections with Tocilizumab treatment

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ABSTRACT

Background

IL-17 has anti-inflammatory activity[24]. IL-17 cytokine production in peripheral blood after *Mycobacterium tuberculosis* infection in human[25] and also COVID-19 infections. This is the first sight to show the extent of the difference in IL-17 concentration levels produced in response to these infections, either before or after therapy.

Methods

TB (forty patients) and the other 40 patients of COVID-19 with healthy subjects were included in the present study to estimate IL-17 using an enzyme-linked immunosorbent assay (ELISA). This study is divided by gender and age group factors, as well as measuring the differences in IL-17 concentrations before and after these diseases, with the therapeutic effect of Tocilizumab in this cases according to the outcome and days of hospital after treatment .

Results

The concentration levels of IL-17 cytokine for tuberculosis and COVID-19 patients according to age group were (234±92.82 and 126.6±15.56 pg/ml, respectively) with a significant statistical difference ($p < .05$), whereas the evaluated value of IL17 for tuberculosis, COVID-19 patients, and control groups according to gender factor was in females (211.6± 65.61, 141.1±35.33 and 86.68±8.15).The results explained that statistics were different between the study groups before and after treatment according to IL-17 concentration levels.

Conclusions

IL-17 appears to be included in immune-mediatory responses in patients with tuberculosis and COVID-19 patients. The study found a significant difference in Iraqi patients for IL-17 concentrations of tuberculosis and COVID-19 infections, with the effect of Actemra on Iraqi patients especially in percent of hospital discharge.

Keywords: IL-17, Tuberculosis, COVID-19, age groups, gender, Actemra

Introduction

More than 2 million people die each year from tuberculosis16, which is caused by the *Mycobacterium tuberculosis*, which also causes cellular illnesses [12.13]. Every body organ, including the liver, lung, and spleen, as well as cells that can respond to adenocytes IL-17, macrophages, lymphocytes, epithelial cells, keratocytes, and fibroids[17, 18], expresses cytokines and their IL-17 (IL- 17RA) receptors[19, 20]. We are constantly learning about the complexity of Th17 cells and Th17-derived cytokines because this is a newly discovered population. Recent research suggests that these various cells and cytokines exert pressure on bacteria, fungus,

and cellular viruses on various mucosal surfaces in a larger and more sophisticated manner. Animal models of autoimmune disorders have demonstrated that Th17 and Th17 cells generated from cytokines are important sources of initiation and tissue damage[14, 15].

As the third national security threat posed by coronavirus 2 respiratory syndrome (SARS-CoV-2) became serious, our study also looked at the levels of the cytokine IL-17 in COVID-19. Fever, cough, fatigue and phlegm production are main clinical symptoms of COVID-19 patients as well as lymphocytopenia and radiological pneumonia evidence [21]. Death Acute respiratory distress, respiratory failure, and sepsis are possible outcomes. Because of multi-organ dysfunction or alveolarinjury from systemic inflammation, cytokines and chemokines have a long period.

It is thought that they are crucial for immunity and immunity ethology during viral infections regardless of declining serum levels. Many cytokines and flammable chemokines have been shared with the severity of the illness and the mortality rate of SARS-CoV-2 were conflicting in these findings conclusions [22, 23]. The weak COVID-19 viruses and the severity-related variables are still unknown.

Actimera contains the important and effective agent tocilizumab), a protein that helps to reduce inflammation by inhibiting the action of cytokins that cause the release of inflammatory substances and interleukins that increase the immune response of cells. [25-26]

Method

Subject

During the period from (December, 2021, to March, 2022), serums were collected from 40 patients with active pulmonary tuberculosis, as well as 40 other serum samples from patients with COVID-19. All samples were checked to show serum IL-17 concentration levels[5]. The patients cheeked to Actemra (tocilizumab) therapy effects for COVID-19.

ELISA

The kit of ELISA for human IL-17 (Quantikine™ ELISA, Bio-Techne China) was used to estimate the concentrations levels of IL-17 following the guidelines provided by the manufacturers.

Statistical analysis

The significance of differences in effects between the groups (Tuberculosis, COVID-19 infected patients and controls of serum IL-17 levels), men, women, and aged groups were determined using the chi-square test and all data were analyzed by version 16 of the SPSS software. Data are expressed on mean ± SD and value below 0.05 ($p < 0.05$ or $p < 0.001$) are different in statistical significance.

Results:

The results of the IL-17 levels of (Tuberculosis, Covid-19) infected patients were compared with the results of the IL-17 concentrations of healthy people. Table (1) shows the differences in the levels of interleukin-17 concentrations with regard to patients with active pulmonary tuberculosis and COVID-19 patients according to the difference in age groups, where the table shows that there are significant differences at $p < 0.05$. Discharge of infected Iraqi patients (COVID-19) from hospital on 21st day of treatment application with (ACTEMRA) (Table 3),

The table also shows that the patients infected with coronavirus who recovered by five to seven days was high when using tocilizumab (44%). On the other hand, no deaths (0%) were seen with the use of tocilizumab treatment.

Table (1): Estimation of IL-17 levels /TB, Covid-19 and control / age groups.

Age group	IL-17 (pg/ml) ofTB patients		IL-17 (pg/ml) of Covid-19 patients		IL-17 (pg/ml) of Control.	
	No.	Mean ±SD	No.	Mean ±SD	No.	Mean ±SD
15-30	11	194.9 ± 41.06	13	134.07±31.75	11	85.9±9.18
31-40	9	196.2 ± 44.32	8	149.15±56.78	8	87.3±7.38
41-50	12	195.7± 52.23	11	156.18±31.65	13	85.9±8.13
51-70	8	234± 92.82	8	126.6±15.56	8	56.12±4.64
P value	significant at $p < .05$					

Table (2): Estimation of IL-17 levels / TB, Covid-19 and control / gender types.

Gender type	IL-17 (pg/ml) ofTB patients		IL-17 (pg/ml) of Covid-19 patients		IL-17 (pg/ml) of Control.	
	NO.	Mean±SD	NO.	Mean±SD	NO.	Mean±SD
females	15	211.6± 65.61	20	141.1±35.33	19	86.6±8.15
males	25	198.28± 56.95	20	139.9±40.5	21	86.4±9.07
P value	not significant at $p < .05$					

Table (2): Day of discharge from hospital and mortality data in covid-19 Iraqi patients.

No.	groups		Actemra		*p-value*
1.	Outcome%	Hospital discharge	40	100%	*>0.05*
	Outcome%	death	0	0%	
2.	Days in hospital after Actemratreatment		3-5 days	0.0	
			5-7 days	45%	
			8-15	68%	
			More than 15 days	100%	

The current study also showed that there are differences in the levels of interleukin with regard to the distribution of the gender of patients, noting that there are no significant statisticaldifferences*, $p < 0.05$* (Table 2).

To find out these differences clearly, these comparisons were made using Mean±SD for active tuberculosis patients and COVID-19 patients, with healthy subjects considered to be in control. This shows that there is a high concentrationof interleukin for tuberculosis patients (203.275 pg/ml) compared to the outcomes of COVID-19 patients (140.5 pg/ml) or control (85.5 pg/ml) asshown in figure (1).

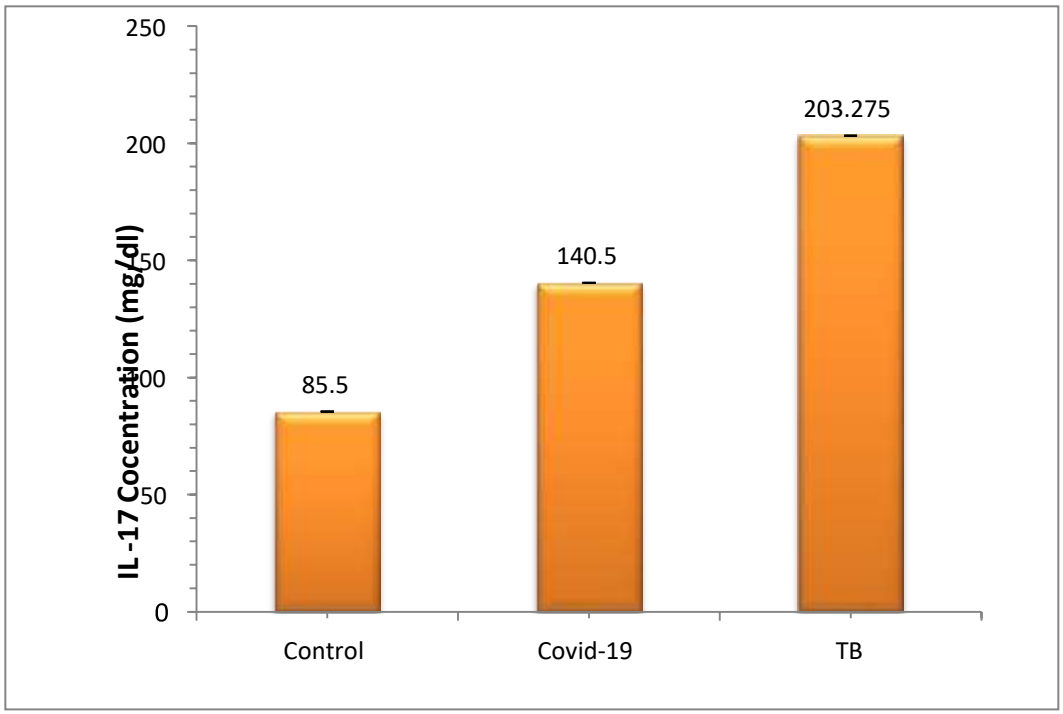

Figure (1): Distribution of IL-17 in TB, COVID-19 patients and control group beforetreatment.

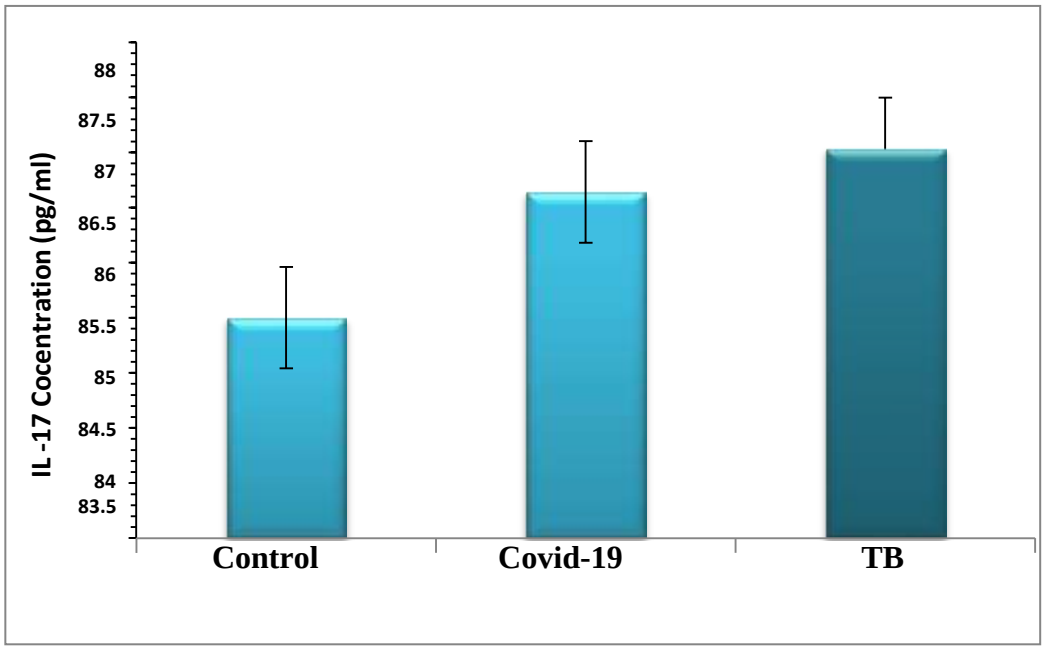

Figure (2): Distribution of IL-17 levels in TB, COVID-19 patients and control group aftertreatment.

In figure (2), it was observed that levels of interleukin in active pulmonary tuberculosis infections had decreased by using first-line anti-drug therapy for four months (except for one relapse of a patient). This decrease was significant enough to reach 87.035pg/ml. On the other hand, the levels of interleukin concentration in COVID-19 patients' infections have been significantly reduced to (86.642 pg/me) after more than 20 days of treatment compared to the result of control (no significant statistical difference).

Discussion

In our current study, we did not find a statistically significant rise in IL-17 concentrations when comparing age and sex for either tuberculosis patients. This comparison is very clear in measurement with the control group (Table 1 and 2). In previous studies, it has been observed that IL-17 may increase in children in a statistically significant way for people with infections[2]. In line with those results, in a recent study of some of the authors, significantly increased levels of IL-17 in different age groups with tuberculosis have not been detected [1,3].

Gopal *et al*. (2014) discovered no statistical significance for the effect of IL-17 cytokin on immunity against hypervirulent tuberculosis in laboratory mice across different factors (gender or age groups).

In our study, the role of IL-17 among COVID-19 patients was also investigated, and there was no statistically significant effect on the factors of age and gender among patients, indicating the rise in the concentration of IL-17 compared to the results of healthy people (control). Some investigations have shown potential roles played by IL-23/IL-17 in COVID-19 infections[18, 19], and these studies have shown that levels of cytokine serum did not have an effect either. Other recent studies have shown that the age and severity of lung involvement also did not affect IL-17 serum levels in corona patients. However, due to our results, the cytokine IL-17 may presumably not play a important role in corona pathogens in Iraqi patients.

Our investigation, we found that the serum IL-17 level was elevated in positive AFB patients (figure 4-1) but later decreased after the AFB test was converted (figure 4-2). Th17, which plays an immunoregulatory role by releasing several proinflammatory cytokines such as TNF-alpha, IL-17A, IL-26, IFN gamma, and IL-22, is primarily responsible for creating IL-177. All of these cytokines stimulate and activate neutrophils, Th1, and macrophages, which aid in the destructionof the infected lung tissue and the slowing of MTB growth [8].

On the other hand, it may be wildly wrong or impossible to understand the intricate role that the cytokine IL-17 plays in immunological processes [9, 10]. However, we may see them with levels of IL-17 concentrations that slightly increase when betacoronaviruses interact with specific immune system machines (as shown in our study, figure 4-1). Due to the novelty of COVID-19 and the sudden onset of these viruses with a limited group of patients with SARS and MERS that have been documented within the studies of many investigators, there is information that it may contribute to immunological disease machines with little data for this virus [6].

In fact, many clinical experiments have been conducted for the purpose of evaluating the effectiveness of tocilizumab, especially in the treatment of pulmonary infections for coronavirus patients, specifically when IL-17 inhibitors with tocilizumab share the same pathways as Th17 signaling and IL-17 suspension factors may be more effective, safe and ability in terms of treatment against COVID-19 and this due to the fact that cytokine (IL-17) may precede of proinflammatory cytokines such as TNF-alpha [26]. The mechanism of administering suspended substances to IL-17 through the subcutaneous way is more efficient than given by intravenous way, with tocilizumab reducing the risk of death in severe pneumonia patients [24].

Conclusion

IL-17/th17 appears to be involved in pro-inflammatory responses in patients with tuberculosis in Iraqi hospitals. However,

gender can be regarded as a risk factor for the production of more cytokines as well as the perceived rise of IL-17 in COVID-19 patients, which needs further exploration.

It is clear that our results have indicated that tocilizumab is an effective inflammatory reducer at the first of treatment. These potential events, especially negative ones, are an important criterion for determining immunosuppressive treatment for many patients as inhibitors of IL-17.

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