Correlation of interleukin-17 with disease activity and hand joint damage in patients with rheumatoid arthritis

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ABSTRACT

Background: Rheumatoid arthritis (RA) is a disease involving various types of cytokines. One of them is interleukin-17 (IL-17), which is known to have a pleiotropic effect on various of cells and is thought to be a cytokine effector contributing to the pathogenic condition in RA.

Methods: The study was conducted on 46 RA patients at rheumatology clinic at the Cipto Mangunkusumo General Hospital who were diagnosed based on the 1987 American College of Rheumatology criteria. Sample selection was done using consecutive sampling. Tests on patients were conducted to collect data needed to obtain the scores for 28-joint Disease Activity Score (DAS28), global health visual analogue scale, swollen joint count, tender joint count, sedimentation rate, Sharp score (radiograph of both hands), and the IL-17 level. The correlation between IL-17 level and DAS28 was calculated using the Pearson's correlation test while the correlation between IL-17 level and Sharp score was calculated using the Spearman's test.

Results: The majority of patients (87%) were women. The largest percentage was in the 51- to 60-year-old group (39.1%). Most patients (43.1%) had moderate disease activity. There were 27 patients (58.69%) with positive rheumatoid factor. The mean IL-17 level was 17.28 pg/mL with a standard deviation of 11.43 pg/mL. There was no correlation of IL-17 level with disease activity ($p = 0.446$, $r = 0.021$) and Sharp score ($p = 0.304$, $r = 0.077$) in subjects of this study.

Conclusion: There was no significant correlation of IL-17 with disease activity and joint damage.

Rheumatoid arthritis (RA) is a systemic autoimmune disorder characterized by polyarthrititis and chronic synovitis resulting in joint damage. Although the pathogenesis of RA is not yet fully understood, it has been proven that the mechanism involves T cells, B cells, macrophage, neutrophil, and synovial fibroblast.[1–4] The various types of cytokines produced by those cells play an important role in the development of inflammation, articular destruction, and other comorbid related to RA. Since the disease activity of RA patients is closely related to changes in levels of those cytokines, the management of RA today involves manipulation of the cytokine tissue.[5–7]

Tumor necrosis factor α (TNFα) is the main cytokine in the pathogenesis of RA. Although inhibition of TNFα had showed good results in RA treatment, there are still cases that did not show good response; thus there must be other cytokines involved that could be used as alternative targets for intervention.[7] One of them is interleukin (IL)-17, the cytokine produced by T cells. Interleukin-17 has a pleiotropic effect on various cells including macrophage and fibroblast and could be a strong candidate for being the cytokine effector that contributes to pathogenic RA. In this study, we examine the correlation of serum IL-17 level with disease activity and Sharp score in RA patients.

METHODS

This is an analytical study using cross-sectional method. The study was performed at the Division of Rheumatology of the Department of Internal Medicine, University of Indonesia School of Medicine/Cipto Mangunkusumo General Hospital, Jakarta from January until February 2010. This study has passed the ethical clearance set by the ethical committee.

The subjects of this study were new and existing RA patients at rheumatology clinic at the Cipto Mangunkusumo General Hospital. The subjects were selected using consecutive sampling. The size of the sample was calculated using the correlation coefficient equation for single sample in which $\alpha = 5\%$ and $\beta = 10\%$, $Z\alpha = 1.96$ and $Z\beta = 1.28$. The correlation coefficient (r) of serum IL-17 level with disease activity and joint damage in hand radiography of RA patients were estimated at 0.5; so the result for the sample size was 37.7 (rounded up to 40).

The inclusion criteria were RA patients who fulfilled the 1987 American College of Rheumatology criteria, older than 16 years during the onset of disease, and willing to participate in the study by signing informed consent. The exclusion criteria were presence of clinical signs of acute infection or other inflammatory process. The patients underwent routine examinations (history taking, physical examination, laboratory tests according to the needs for data collection) and finally radiography of both hands. The data collected were processed using Statistical Package for the Social Sciences (SPSS) program.

Interleukin-17 is a cytokine present in the circulation of RA patients. IL-17 level was quantified using enzyme-linked immunosorbent assay and

measured in pg/mL. The reagent used was Quantikine Human IL-17 Immunoassay (R and D System Inc., Minneapolis, USA). The measurement scale is numeric.

Disease activity was measured using 28-joint Disease Activity Score (DAS28)[8,9] with four variables: swollen joint count (SJC, total 28), tender joint count (TJC, total 28), sedimentation rate (SR), and global health assessment using visual analogue scale (VAS) with a scale ranging from 0 to 100. The measurement scale is numeric.

Joint damage in hand radiograph was measured using Sharp score[10,11] and obtained from the total score of joint space narrowing in 16 areas and bone erosion in 17 areas of the radiographic image of the right and left hands. Hand radiographs were taken in the anterior-posterior position using the device from Toshiba KXO-15E, DRYFIX 7000, Capsula FCR XLII. The measurement scale is numeric.

RESULTS

Characteristics of subjects

Of the 46 subjects involved in this study, 40 patients were women (87%) and 6 were men (13%). Their ethnic background and origins varied: Javanese 15 patients (32.6%), Sundanese 12 (26.1%), Batak 7 patients (15.3%), Betawi 3 patients (6.6%), Minang 3 patients (6.6%), Lampung origin 2 (4.5%), and one patient of each of Dayak, Malay, and Aceh origin (2.2%).

Mean age of the subjects was 48.6 years old, mostly were in the 51- to 60-year-old age group. The distribution of the age group were as follows: 1 patient (2.2%) was in the 21- to 30-year-old group, 8 patients (17.4%) in 31- to 40-year-old group, 15 patients (32.6%) in 41- to 50-year-old group, 18 patients (39.1%) in 51- to 60-year-old group, and 4 patients (8.7%) in 61- to 70-year-old group.

The outcome of the disease activity score based on DAS28 were as follows: 13 patients (28.3%) had high activity (DAS28 >5.1), 19 patients (41.3%) had moderate activity (3.2<DAS28≤5.1), 8 patients (17.4%) had low activity (2.6<DAS28≤3.2), and 6 patients (13%) had remission (DAS28 ≤2.6).

Twenty seven (58.69%) patients had positive rheumatoid factor while 19 patients (41.31%) had negative rheumatoid factor.

Other characteristic data obtained such as the tender and swollen joint counts, global health VAS score, DAS28 score, IL-17 level, and Sharp score are presented in table 1 while the distribution of serum IL-17 levels of the RA patients according to the disease activity are presented in table 2.

Table 1 Characteristics of subjects

Characteristics	Mean (SD)	Median (range)
Age, years	48.61 (9.23)	50 (29–67)
Tender joint count	5.41 (6.28)	3 (0–22)
Swollen joint count	2.54 (3.67)	0 (0–14)
Global health VAS	17.50 (17.76)	10 (0–70)
Sedimentation rate, mm/hr	58.50 (32.10)	52.5 (3–116)
DAS28 score	4.24 (1.37)	4.30 (1.74–7.46)
Joint space narrowing score	5.41 (6.22)	3 (0–21)
Bone erosion score	3.48 (4.71)	2 (0–18)
Sharp score	8.89 (9.87)	5 (0–34)
Interleukin-17 level, pg/mL	17.28 (11.43)	13.44 (8.11–73.23)

VAS, visual analog scale; DAS28, 28-joint Disease Activity Score.

Table 2 Distribution of serum interleukin-17 (IL-17) level of RA patients according to the disease activity

Disease activity	N	IL-17 level, pg/mL	
		Mean (SD)	Range
Remission	6	14.03 (4.33)	9.83–20.68
Active	40	17.77 (12.10)	8.11–73.23

Correlation between IL-17 level and disease activity

Serum IL-17 level tests were conducted on the 46 patients. The mean level obtained was 17.28 pg/mL with a standard deviation of 11.43 pg/mL. Normality test of the transformed IL-17 data and DAS28 showed a normal distribution; hence Pearson's correlation test was used. Although IL-17 level was assumed to fluctuate in the serum according to disease activity, the outcome of the Pearson's test in this study showed that there was no significant correlation between IL-17 level and disease activity measured with DAS28 ($p = 0.446$, $r = 0.021$). The scatter diagram of the correlation between IL-17 level and DAS28 is presented in figure 1.

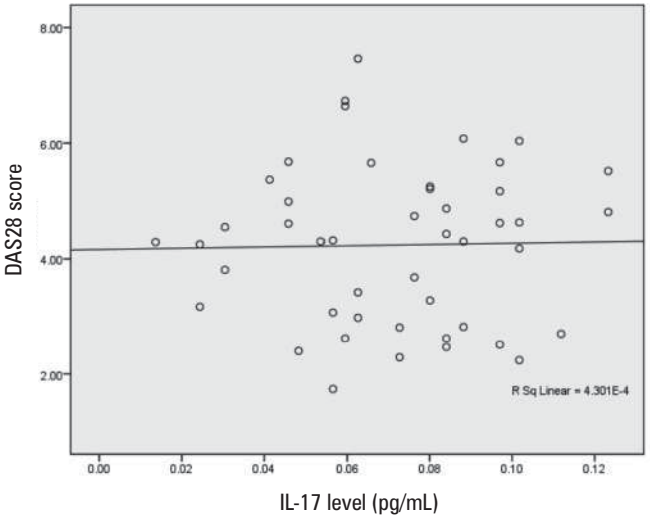

Figure 1 Correlation between interleukin-17 (IL-17) level and 28-joint Disease Activity Score (DAS28).

Correlation between IL-17 level and hand joint damage

Hand joint damage in this study was measured using Sharp score that consists of joint space narrowing and bone erosion score based on the radiographic examination.

Correlation between IL-17 level and joint space narrowing score

Normality test revealed that the variable data of joint space narrowing had an abnormal distribution; so nonparametric statistics was used for the bivariate analysis. The outcome of the Spearman's test between IL-17 level and joint space narrowing in hand radiograph showed no significant correlation ($p = 0.406$, $r = 0.036$). Figure 2 shows the scatter diagram of the correlation between IL-17 level and joint space narrowing score.

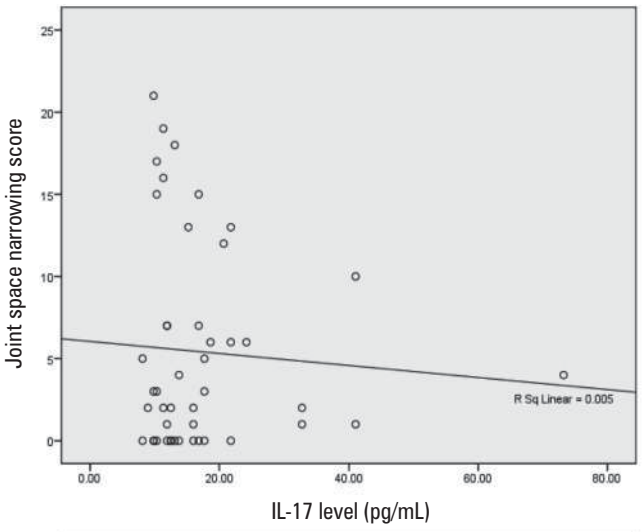

Figure 2 Correlation between interleukin-17 (IL-17) level and joint space narrowing score.

Correlation between IL-17 level and bone erosion score

The normality test of the variable data of bone erosion showed an abnormal distribution; so nonparametric statistics was used for the bivariate analysis. The correlation between IL-17 level and bone erosion showed a trend in which the higher the IL-17 level, the greater the erosion becomes. However, statistical test using Spearman's test failed to show a significant correlation ($p = 0.181$, $r = 0.138$). The scatter diagram of the correlation between IL-17 level and bone erosion score is shown in figure 3.

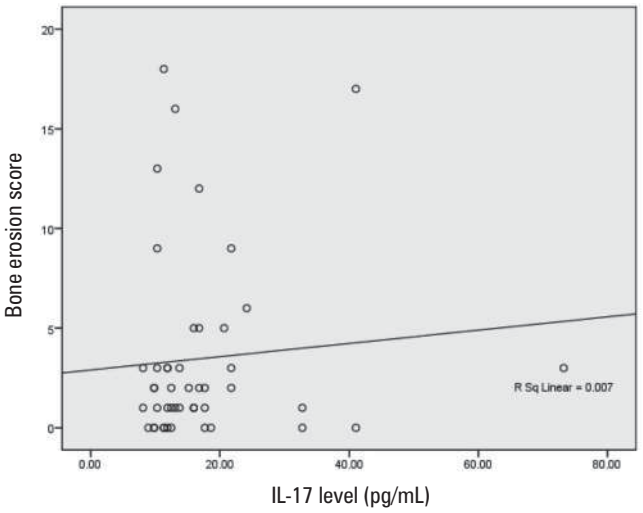

Figure 3 Correlation between interleukin-17 (IL-17) level and bone erosion score.

Correlation between IL-17 level and Sharp score

The Sharp score is the total value of the joint space narrowing and bone erosion in hand radiograph. The normality test of variable data of the Sharp score showed an abnormal distribution; so nonparametric statistics was used for the bivariate analysis. The outcome of the Spearman's test between IL-17 level and Sharp score showed no significant correlation ($p = 0.304$, $r = 0.077$). The scatter diagram showing the correlation between IL-17 level and the Sharp score is presented in figure 4.

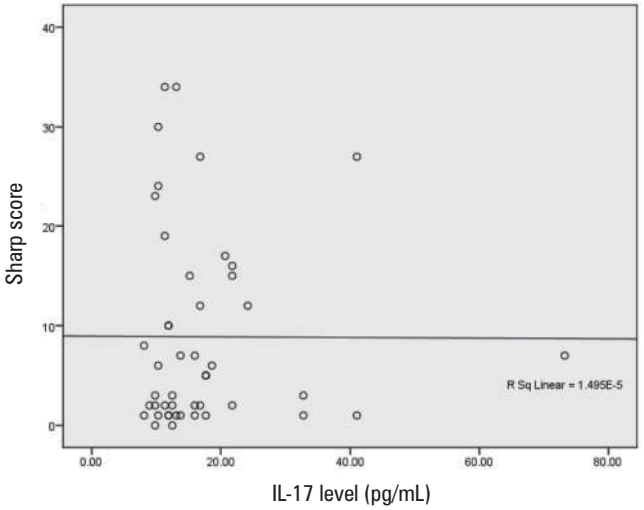

Figure 4 Correlation between interleukin-17 (IL-17) level and Sharp score.

DISCUSSION

In recent years there are increasing numbers of RA cases found, which may likely be related to the increased awareness of this disease. In the general population, the prevalence of RA is higher in women and increases with age. Approximately 0.12% of patients was in the age group of 16 to 44 years old and rises to 2.99% in the above 75-year-old age group.[12] In this study we found more female patients (87%) than male patients with a ratio of 6.7 to 1. The subjects were from different ethnic groups of Indonesia but were not representative of the various ethnic groups spread across Indonesia. The ethnic factor is reported to influence the prognosis of RA. The subjects in this study were mostly in the 51- to 60-year-old age group; however, the tendency of RA prevalence in Indonesia is in the younger age group. The reason for this different finding is still not clear.

The mean serum IL-17 level of the subjects was 17.28 pg/mL with a range of 8.11–73.23 pg/mL. It is higher than the level found in healthy individuals in other studies: 0.28–5.47 pg/mL as reported by Ciprandi et al,[13] 0.0–13.4 pg/ml with a mean of 7.4 pg/mL by Arican et al,[14] and a mean of 0.01±0.0 pg/mL by Hussein et al.[15] The IL-17 levels found in this study were also higher than those in psoriatic arthritis patients.[14] However, due to the lack of control group we cannot yet draw a conclusion whether IL-17 level in the subjects of our study was higher than healthy individuals in our population. Regarding this issue, we are planning to publish an extended report of this study with inclusion of a control group.

Table 3 Interleukin-17 levels (pg/mL) in healthy people, arthritis, and allergy

	Healthy	Arthritis	Allergy
Ciprandi et al[13]	0.28–5.47	-	1.81–141.29
Arican et al[14]	0.0–13.4	8.3±3.8*	-
Hussein et al[15]	0.01±0.0	0.2±0.1**	-
This study	-	8.11–73.23	-

*psoriatic arthritis; **rheumatoid arthritis.

Based on the disease activity, only 6 patients (13%) had remission while most patients (41.3%) had moderate activity. The cause of low remission rate among patients in this study—either suboptimal treatment or others—is still open for further studies.

Routine clinical follow-ups of RA patients should be performed by their personal doctors. Evaluation of disease activity score is important in the follow-ups, since the result will have an effect on the disease management. As one of the inflammatory cytokines that played role in the pathogenesis of RA, fluctuations of IL-17 concentration will influence disease activity. Shahrara et al[16] reported an increased number of Th17 cells found in the synovial fluid and peripheral blood of RA patients. Hussein et al[15] reported higher levels of serum IL-17 in RA patients than those in healthy control subjects. In addition, Anderson et al[2] reported high expression of IL-17 in the synovial tissue and circulation. Romagnani[17] also explained the presence of high IL-17 level in the serum that plays a role in the process of recruitment, activation, and migration of neutrophils and stimulates production of IL-22, which greatly increases in chronic inflammation. The involvement of the above cytokine will cause migration of inflammatory cells and an increase in immune complex formation. These conditions will clearly result in an increase in disease activity.

We found no significant correlation between serum IL-17 level and disease activity ($p = 0.446$, $r = 0.021$) in this study. There are several possible explanation for this finding. First, IL-17 may not be the dominant cytokine in the subjects. Second, it has to act through certain mediator and thus has only indirect effect on the disease activity. Further studies using other methods are needed to determine the actual role of IL-17 and its relation to disease activity. The outcome in this study was similar to those of Yamada et al[18] and Zrioual et al,[19] who also found no significant correlation between IL-17 and DAS28.

It is clear from the distribution of IL-17 level according to disease activity that patients in remission or active state have higher mean levels than healthy people with a tendency that IL-17 levels are higher in active RA. Despite this finding, we found that the two groups showed no significant difference ($p = 0.29$).

Hand joint damage in this study was evaluated using Sharp score, a radiographic examination to evaluate two conditions: joint space narrowing and bone erosion score. Narrowing of joint space (which occurs primarily as a result of the deterioration of the joint cartilage) and bone erosion are irreversible conditions, the process of which occurs during a certain period of time.

Cytokine synthesis is a cellular activity mediated by the messenger ribonucleic acid (mRNA), an unstable molecule that causes the cytokine synthesis to be a transient process. The involvement of cytokine in RA pathogenesis, mainly in synovitis, is characterized by the migration of inflammatory and immune cells to the synovial tissue. Besides local effect, cytokine also causes systemic effect, especially in acute attacks, with severe inflammation and constitutional symptoms of fever, weakness, fatigue, and muscle pain. Despite this, chronic inflammatory process seems to be dominant in the joint space and tissue.[20]

Interleukin-17 will bind to the IL-17 receptor and is expressed in the epithelial cell, endothelial cell and fibroblast, then it will trigger transcription activation of nuclear factor-κB (NF-κB) and protein-38, which is a mitogen-activated protein kinase, and finally will stimulate secretion of IL-1, TNFα, IL-6, IL-8 or prostaglandin E2; therefore, IL-17 has an important role in immune regulation and inflammatory response, tissue homeostasis, and progression of autoimmune disease. In addition, IL-17 also has a synergic effect with TNFα and IL-1 in inducing joint inflammation and destruction of bone and joint cartilage.[21,22]

From the analysis of this study, we could not obtain enough evidence to show statistically significant correlations of serum IL-17 level with joint space narrowing, bone erosion, and Sharp score. Correlation between IL-17 and hand joint damage could possibly be proven if the tests were conducted in serial. Moreover, the subjects were not in the acute phase. Local inflammation in the form of synovitis was dominant in the subjects even when they were not in remission, which could possibly be caused by the indirect role of IL-17 in causing bone and cartilage destruction. In addition, Moran et al[23] reported that the joints of RA patients during an inflammatory state also produces IL-17 locally, resulting in joint cartilage damage, especially with presence of other cytokines such as TNFα and oncostatin M.

CONCLUSIONS

The mean serum IL-17 level in RA patients in this study was 17.28 pg/mL. A more definitive conclusion regarding IL-17 level in our population may be achieved by the inclusion of a control group. The serum IL-17 levels in active RA patients were higher than those of patients in remission. The serum IL-17 level of RA patients in this study was not significantly correlated with disease activity (DAS28) and hand joint damage (Sharp score). The actual role of IL-17 in the pathogenesis of RA is still to be proven in further studies.

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