Interleukin 6 Concentration in Synovial Fluid of Patients with Inflammatory and Degenerative Arthritis

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Abstract: Aim: The present study aims to compare interleukin 6 concentration in synovial fluid in patients with known types of arthritis.

Background: Persistent synovitis without known markers, such as Rheumatoid Factor (RF), Anti-Citrullinated Protein Antibodies (ACPA), and genetic markers as HLA-B27, is not uncommon. It is valuable to determine the presence of chronic inflammation and put it in correlation with age-related changes, which are especially relevant for middle-aged patients with mono- or oligoarthritis, when the dilemma to start disease-modifying drugs for inflammatory disease often is present. Interleukin 6 (IL-6) plays a significant role in chronic inflammation.

Objectives: IL-6 concentration in synovial fluid reflects the presence and activity of joint inflammation.

Methods: Synovial fluid was obtained from 101 patients with chronic synovitis. IL-6 concentration was determined by the immunochemical luminescence method.

Results: The median IL-6 concentration in synovial fluid in patients with osteoarthritis (OA) was 138.0 pg/ml (interquartile range (IQR) 43.4 to 296.0); in patients with rheumatoid arthritis was 2516.5 pg/ml, (IQR 1136.0 to 25058.0); in reactive arthritis 2281.0 pg/ml (IQR 1392.0 to 8652.0); psoriatic arthritis 1964.0 pg/ml (IQR 754.0 to 7300.0); ankylosing spondylitis 2776.0 pg/ml (IQR 514.7, 3944.0); in a group with negative RF, ACPA and HLA-B27 inflammatory arthritis 2163.0 pg/ml (IQR 822.0 to 7875.0). There is statistically significant difference of IL-6 concentration comparing OA and each inflammatory arthritis group, p<0.0001.

Conclusion: IL-6 detection in the synovial fluid is helpful in arthritis evaluation. The results show that an IL-6 level over 1000 pg/ml suggests the diagnosis of inflammatory arthritis.

Keywords: Interleukin 6, synovial fluid, synovitis, inflammation, inflammatory arthritis, osteoarthritis.

1. INTRODUCTION

Interleukin 6 (IL-6) is a known pleiotropic proinflammatory cytokine, which plays a significant role in chronic inflammation and autoimmunity. IL-6 induces vascular endothelial growth factor activation and angiogenesis in the synovium, increasing vascular permeability resulting in synovitis. It promotes receptor activator of NF-kB ligand activation and osteoclast differentiation leading to bone resorption and induces matrix metalloproteinases production with cartilage degeneration. In the initial stage of inflammation, IL-6 is produced in the local lesion. Onwards IL-6 disseminates via the bloodstream, causing other pleiotropic effects like induction of acute-phase protein synthesis, Th17 cells differentiation, autoantibody production, thrombocytosis, etc. [1].

The serum level of IL-6 does not reflect the presence and activity of joint inflammation. Some studies show low or undetectable serum IL-6 levels and no correlation with IL-6 concentration in synovial fluid [2, 3]. However, the IL-6 level in synovial fluid is significantly higher than the serum level in rheumatic disease patient tests [2, 4, 5].

The present study aims to compare the concentration of IL-6 in synovial fluid in patients with the diagnosed type of arthritis.

2. MATERIALS AND METHODS

The synovial fluid samples were collected during routine joint arthrocentesis for therapeutic reasons from 60 females and 41 males, mean age 47.8 years (the youngest patient 12 years old, the oldest 79), with a known type of chronic arthritis, mostly from knee joints. Clinical diagnoses were confirmed prior by the history, clinical presentations, and specif-
ic radiological investigations in accordance with the current classification criteria for the specific disease. All patients were being treated with nonsteroidal anti-inflammatory drugs, patients with inflammatory arthritis with disease-modifying antirheumatic drugs such as methotrexate, sulfa-lazilone, leflunomide, hydroxychloroquine, and steroids.

Osteoarthritis (OA) had 36 patients and 65 patients - inflammatory arthropathies: 8 patients with rheumatoid arthritis (RA), 54 with seronegative spondyloarthropathies (4 ankylosing spondylitis, 13 psoriatic arthritis, 37 reactive arthritis), and 3 with juvenile idiopathic arthritis. The local ethics committee approved the study. The IL-6 quantitative measurement was made by the immunochemical luminescence method using the immulite 2000 Systems Analyzers according to the manufacturer's instructions. The descriptive statistics method was used for data analysis. The Kruskal-Wallis test was used for IL-6 concentration pairwise comparison in the different diagnosis groups.

3. RESULTS

We analyzed the synovial fluid from 101 patients. Detectable levels of IL-6 (> 1 pg/ml) were observed in all patients.

The median IL-6 concentration in synovial fluid in patients with osteoarthritis (OA) was 138.0 pg/ml (interquartile range (IQR) 43.4 to 296.0) without any significant differences in primary and secondary osteoarthritis. In inflammatory arthropathies, the median IL-6 level was 2194.0 pg/ml (IQR 1254.0 to 8625.0). Median IL-6 concentration in patients with RA was 2516.5 pg/ml (IQR 1136.0 to 25058.0). There was no significant difference between patients with seropositive and seronegative RA groups. In reactive arthritis median IL-6 level was 2281.0 pg/ml (IQR 1392.0 to 8652.0); psoriatic arthritis - 1964.0 pg/ml (IQR 754.0 to 7300.0) and ankylosing spondylitis - 2776.0 pg/ml (IQR 514.7, 3944.0). In a group of 19 patients with negative rheumatoid factor (RF), anti-citrullinated protein antibodies (ACPA) and HLA-B27, median IL-6 level in synovial fluid was 2281.0 pg/ml (IQR 1392.0 to 8652.0); psoriatic arthritis - 1964.0 pg/ml (IQR 754.0 to 7300.0) and ankylosing spondylitis - 2776.0 pg/ml (IQR 514.7, 3944.0). In a group of 19 patients with negative rheumatoid factor (RF), anti-citrullinated protein antibodies (ACPA) and HLA-B27, median IL-6 level in synovial fluid was 2281.0 pg/ml (IQR 1392.0 to 8652.0). In inflammatory arthritis with negative immunological and genetic markers such as RF, ACPA, and HLA-B27. For most cases, the IL-6 concentration in synovial fluid was more than 1000.0 pg/ml. In the case of OA, there was also IL-6 elevation, however, it was less elevated with a range of 43.4 to 296.0 pg/ml.

4. DISCUSSION

IL-6 concentration in synovial fluid tested in patients with different types of arthritis, including inflammatory arthropathies, OA, and crystal deposition diseases. IL-6 level in the synovial fluid of patients with inflammatory arthropathies [4, 6] is significantly higher than that of patients with osteoarthritis [5, 7, 8].

In comparison to healthy individuals, IL-6 concentration is increased in the synovial fluid of OA patients and patients with symptomatic cartilage defects [9, 10].

RA studies show that IL-6 concentration in synovial fluid correlates with synovial tissues inflammatory cells infiltration, synovial fluid hypercellularity [5, 11], histological characteristics of chronic synovitis [3], and with radiologic stages of joint destruction [12]. OA studies show that IL-6 level in synovial fluid correlates with the presence of cartilage degrading matrix metalloproteinases in synovial fluid and radiographic disease severity [2].

In a knee joint injury, IL-6 concentration in synovial fluid is highly elevated compared to normal controls, and IL-6 level in synovial fluid is higher during the first 24 h after injury and decreases afterward [13, 14]. Comparing end-stage OA and joint injury groups shows elevated IL-6 concentration in the OA group [15].

This study observed elevated IL-6 concentration in synovial fluid in patients with inflammatory arthritis with no differences between patients with seropositive and seronegative RA. High IL-6 concentration in synovial fluid was observed in inflammatory arthritis with negative immunological and genetic markers such as RF, ACPA, and HLA-B27. For most cases, the IL-6 concentration in synovial fluid was more than 1000.0 pg/ml. In the case of OA, there was also IL-6 elevation, however, it was less elevated with a range of 43.4 to 296.0 pg/ml.

Fig. (1). IL-6 concentration in patients with OA. OA, osteoarthritis; n, number of patients.

Fig. (2). IL-6 concentration in patients with inflammatory arthritis. RA, rheumatoid arthritis; ReA, reactive arthritis; PsA, psoriatic arthritis; n, number of patients.
There is not much research dedicated to IL-6 concentration in synovial fluid and its correlation with rheumatic diseases. Overall sample numbers in the case of inflammatory arthritis are low. The presented data is not directly comparable because of the differences in patients groups, methods of IL-6 detection, and data presentation. However, the data is consistent with that of other researchers (Table 1). In general, the presented data show the tendency of more pronounced elevation of the IL-6 concentration in the synovial fluid in case of inflammatory arthritis and is statistically significant in the case of inflammatory seronegative arthritis. But the slight elevation of IL-6 concentration, usually not exceeding 500 pg/mL, is in OA.

Chronic persistent mono-/oligo-arthritis is not uncommon in clinical practice, especially in the knee joint. Progression of inflammatory arthritis leads to secondary OA. But there are currently no markers that can help to distinguish joint inflammation from degeneration in some circumstances, such as inconclusive blood tests when inflammatory markers, immunological and genetic parameters are absent. Radiological investigations are helpful to determine the extent of damage caused by arthritis, however they are not always beneficial to distinguish when treatment is indicated. Therefore, measurement of IL-6 concentration in synovial fluid may be an option. Having the IL-6 concentration is valuable to determine chronic inflammation and differentiate it from age-related changes, which is especially relevant for middle-aged patients with mono- or oligoarthritis when there is a dilemma for treatment with disease-modifying drugs for inflammatory disease.

### CONCLUSION

The study shows that an IL-6 concentration in synovial fluid over 1000 pg/mL consistent with the diagnosis of inflammatory arthritis. Based on the research results, the IL-6 testing is worth being implemented in clinical practice more widely as this may be helpful as an additional tool in the diagnostic process.

### ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The Ethics Committee of Riga Stradiņš University in Latvia approved the study protocol (Nr. 6-1/05/26).

### HUMAN AND ANIMAL RIGHTS

No animals were used in this study. The reported experiments on humans were followed in accordance with the Helsinki Declaration of 1975, as revised in 2013.
CONSENT FOR PUBLICATION

All individuals participating in this study provided informed consent.

STANDARDS OF REPORTING

STROBE guidelines were followed.

AVAILABILITY OF DATA AND MATERIALS

The data that support the findings of this study are available from the corresponding author [AM], upon reasonable request.

FUNDING

None.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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