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The Relationship Between Some Serum And Synovial Fluid Cytokines And Pain Level At The Early Stage Of Knee Osteoarthritis

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Abstracts

Background: Some cytokines such as IL-1β, TNF-α, VEGF, IL-6, IL-15, IL-17, IL-18, IL-4, IL-10, and IL-13 had been found in the knee joint and demonstrated the correlation to knee injuries and clinical symptoms. Paint is the most important symptom in OA. However, the causes and correlative factors of paint in OA are still not clear.

The main aim: To study the determination IL-1b, IL-6, IL8, and IL4 in serum and synovial fluid in the knee of osteoarthritis patients at the early stages and their relationship to pain level.

Materials and Methods: A cross-sectional descriptive study was conducted in 35 knee osteoarthritis patients with knee effusion from April 2018 to July 2019 in Military Hospital 103. The serum and synovial fluid were used to determine the cytokines level and to estimate the correlation between them and some clinical features.

Results: There were no significant differences between the cytokines levels in terms of age, gender, pain level as the visual analog scale score, and the duration of disease except IL4 in synovial fluid were differences between the visual analog scale score group and osteoarthritis stages. The interleukin levels in synovial fluid had a high correlation to each other. There was no significant correlation between their levels in the serum.

Conclusion: In synovial fluid, only the IL4 concentration was significantly different between osteoarthritis stage and pain level groups.

Keywords: cytokine, interleukin, osteoarthritis, knee, pain level, synovial fluid.

1. Introduction

Osteoarthritis (OA) appears in hip and knee might mainly cause pain and hard exercise. Those effects on many people quality and associate to premature mortality [1]. One more important thing should be said: osteoarthritis is a disease with a severe course and large term for rehabilitation, and one of all becomes the restoration of the gait [2].

The Global Burden of Disease study in 2017 showed that approximately 263 million people (equivalent to 63.7 % population) had OA of the knee [3]. In the joint tissue, the damage and regeneration are two parallel processing. Although the radiographic images were the criteria for OA diagnosis, the changes in joint tissues appeared for a long time before. Images could not reflect the biochemical changes happening in the joints. Even though OA is regarded as a non-inflammatory system disease, many inflammatory cytokines had important roles in the OA process such as IL-1β, TNF-α, VEGF, IL-6, IL-15, IL-17, IL-18, IL-4, IL-10, and IL-13 [4-11]. These cytokines had been found in the knee joint and demonstrated the correlation to knee injuries and clinical symptoms. Paint is the most important symptom in OA. However, the causes and correlative factors of paint in OA are still not clearly [12-13]. Evaluation of these cytokines could show not only their relationship in OA pathology more clearly but also their correlation to some features.

2. Materials and Methods

The cross-sectional study was conducted in 35 patients who had been diagnosed OA of the knee with effusion at the Military Hospital 103, Hanoi, Vietnam, from April 2018 to July 2019. OA patients were diagnosed by the American College of Rheumatology criteria for the diagnosis of knee osteoarthritis[12]. Research participants have not included patients having history of trauma, diseases of rheumatoid arthritis, previous joint infection, crystal deposition arthritis, enteropathic arthritis, hemophilic

arthropathy, systemic inflammatory or autoimmune disorders, malignant disease, advanced renal disease, Diabetes Mellitus, histories of corticosteroids medication, or refused to participate in the study (Table 1).

2.1. Pain level

The level of pain was evaluated on a visual analog scale (VAS). This scale appeared on a two-sided ruler. One side was divided from 0 to 100 (equivalent to 100 mm). The other side was marked at both ends of the line corresponding to the straight line on the front: painless corresponds to the number 0; the most pain level imaginable is 100. When evaluating the pain level, the patient could only see the back (no number) and self-assess the pain level, drag the ruler to the degree of pain self-assessment. The physician would refer to the number on the front of the ruler to determine the degree of pain from 0 to 10.

Pain measurement using a VAS scale:

0 – 10	: No pain
10 – 40	: Mild pain
40 – 70	: Moderate pain
70 – 100	: Severe pain

2.2. Osteoarthritis stage

This classification was proposed by Kellgren and Lawrence in 1957, and later accepted by the World Health Organization (WHO) in 1961 as the radiological definition of OA for the purpose of epidemiological studies.

Disease severity in patients with OA of knee was determined from radiographs. Five OA grade divisions (0-4) were assigned based on radiographic analysis.

+ Grade 0: No radiographic features of OA are present.
+ Grade 1: Doubtful joint space narrow (JSN) and possible osteophytic lipping.
+ Grade 2: Definite osteophytes and possible JSN on anteroposterior weight-bearing radiograph.
+ Grade 3: Multiple osteophytes, definite JSN, sclerosis, possible bony deformity
+ Grade 4: Large osteophytes, marked JSN, severe sclerosis and definite bony deformity.

All the relevant information was provided in supplementary data.

2.3. Samples collections

Blood was collected from patients in the morning. After centrifugation, serum was collected and stored at -80^{o}C for subsequent analysis.

Synovial fluid (SF) was aspirated from the knee joint by FNA (Fine Needle Aspiration) via ultrasound method. The SF could only be collected from the patients who had feeling of heaviness, puffier look in the knee joint due to ethical reasons. About 2 ml of SF was collected into an Heparin tube, then centrifured about 4000 prm, and stored at -80^{o}C for measurement. Time from sampling until placed in the freezer ranged from 2 to 4 hours.

2.4. Multiplex arrays

The reagent used in this study was Human 13-Plex Human ProcartaPlex™ Panel 2, Platinum (Thermofiser, USA). All serums and SF were used to determined four cytokines IL-1β, IL-4, IL-6 and IL-8 according to the manufacture's instruction. The reagents were based on the sandwich immunology principle.

2.5. Ethical statement

This study was approved by the Ethical Committees of Vietnam Military Medical University (Reference No.110/2018/IRB-VMMU). All patients provided written informed consent, and this study was conducted in accordance with the Declaration of Helsinki.

2.6. Statistics

The data were analyzed in Microsoft Excel 2013 and Spss 20.0. Quantitative characteristics were presented in mean, standard division, median. Means of two groups were compared by independent sample t test. The correlation among the variables was accessed by Pearson correlation test. The results were considered to be significant at $p<0.05$.

3. Results

3.1. Cytokine levels in serum and synovial fluid

The IL-1β levels in serum and SF were 1.34 ± 5.31 pg/ml and 0.76 ± 2.44 pg/ml respectively. The IL-8 levels in serum and synovial fluid were 19.2 ± 44.5 pg/ml and 82.63 ± 234.78 pg/ml respectively. The IL-6 concentration in serum and SF was 4.43 ± 3.97 pg/ml and 609.54 ± 929.26 pg/ml respectively. The IL-4

level in serum and SF were 3.24 ± 8.61 pg/ml and 5.82 ± 12.19 pg/ml respectively. Only IL-6 levels in serum and SF were different significantly with $p< 0.05$. (Figure 1)

The IL-4 concentration in SF between two VAS score groups (mild and moderate levels) and between two stages (stage 2 and 3) were different (p significantly). No significant differences of others cytokine levels between duration of disease, BMI groups, pain level groups as VAS score, OA stages (Table 2)

In the synovial fluid, IL-8, IL-6 and IL-1β correlated to each other (p significantly) except IL-4 were not correlation to three cytokines. In serum, they had significant high correlation to each other (Table 3).

4. Discussion

In our study, the level of cytokines that had bad effects and protective effects on cartilage were determined at the same time in both serum and synovial fluid. In the serum, the concentration of all cytokines was not increased significantly. Many researches with similar results had suggested that osteoarthritis had not been a classic arthritis pathology [10,14,15]. In normal people, the joint is a virtual space containing a layer of synovial fluid. In synovial fluid, there is Hyaluronic acid (HA) and some substances permeated from the synovial membrane capillary to act as a lubricant and nourish cartilage. In contrast, in patients with osteoarthritis, HA was lower and less effective in terms of lubrication. This had made the damage of cartilage more severe, broken cartilage structure and appeared foreign objects in the joint, thereby stimulated inflammatory response. Synovitis might be the result of either an acute "flare" of inflammation or chronic inflammation. Inflammation is usually a secondary result, possibly from the accumulation of degradation products of cartilage tissue in the joint [15-18]. In OA patients with inflammatory response, clinical manifested some of the following symptoms: joint swelling, effusion and stiffness. OA was not considered a classic arthritis disease, because there were no neutrophilic granulocytes in synovial fluid, and there was no manifestation of systemic inflammation [15,17,19]. However, synovitis was still common in both the early and late stages of OA. Pre-inflammatory cytokines had been observed in osteoarthritis diseases at an early stage, and synovitis was common in the later stages of osteoarthritis, with involvement of activated T lymphocytes infiltrate into the synovial membrane of the joint. It seems that the levels of the catabolic enzymes and the inflammatory mediators (such as prostaglandin and nitric oxide in synovial fluid and the tissues of

degenerated joints) were positively correlated with the concentration of cytokines like IL-1, IL-6 [11,16,20].

The level of these cytokines had a strong correlation to each other in SF. Although SF was not an inflammation fluid, there was the amount of cytokines that participated in the balance of cartilage degradation and synthesis. Each of them had its role in the OA processing. Interleukin-1 (IL-1) was released by the fibroblast and macrophage-like cells of the synovium in response to cartilage breakdown. IL-1 is a key cytokine in direct others cytokines in anti-inflammatory. IL-1β induced the expression of MMPs, aggrecanases, active plasminogen factors and other pro-inflammatory cytokines such as IL-6, IL-8, leukemia inhibitory factors, IL-17, IL-18 and chemokines. IL-1 stimulated producing more nitric oxide, cyclooxygenase 2 (COX-2), and prostaglandin E2, which contributed to articular inflammation and cartilage destruction. In pathophysiology of OA, IL-1β and TNF-α were considered conductor cytokines, they determine the concentration of other cytokines in this process [6,8,18,21-24]. Many pro-inflammatory cytokines derived from articular cartilage cells such as IL-6 induced the production of protein hydrolyzate enzymes and promoting the progression of osteoarthritis. IL-6 caused inflammation and inhibited regeneration articular cartilage. The production of IL-6 in joint tissues was affected by the response to IL-1β and TNF - α and was carried out primarily by cartilage, osteoblasts, macrophages, lipo cells [14,25-26]. Meanwhile, IL-4 reduced inflammation and enhanced cartilage regeneration. IL8 was produced most by macrophages and had two primary functions. It induced macrophage migration and promoted angiogenesis [26-27]. In our results, only IL-4 level in serum and SF were significant different between groups of VAS score and OA stage. That means the role of IL-4 in reduction knee paint and protection the cartilage. Our results were similar to others [7,26,28]. Understanding the relationship between cytokines and clinical factors plays an important role in detecting and preventing the progression of osteoarthritis [14,18,29].

IL-1β and IL-6 had the promoting role in the inflammatory and destroying cartilage. In our study and many others, their concentration increased at the early stage [30-32]. Although the correlation between pain level (measured by VAS score) and interleukin levels was not demonstrated in this study, some research had shown a weak to moderate correlation between them [26]. There was no significant correlation between interleukin levels to the age, gender, OA stage. These results were similar to others [7,10,26,28].

The first limitation of this study was the cross-sectional design and concentration qualify only. Therefore, the relationship between these interleukins and their role in the disease could not be

clarified and demonstrated. And secondly, the number of patients was not in all 4 stages of diseases. Our study only included patients in stage 2 and 3. So this was not enough information on the earlier and late stage. The third limitation was the causes and risks of OA that had not been clear. So, that the relationship between the fluid appearance in knee joints and the interleukin levels was not found. Furthermore, our study could not prove the part of each cytokine in a general pathway that reflected the relationship between them in disease process.

5. Conclusions

The IL-1β, IL-4, IL-6 and IL-8 had a strict correlation to each other in the synovial fluid in OA patients. In synovial fluid, the IL4 concentration were significantly different between OA stage and pain level groups.

5.1. Clinical significance

- ✓ The cytokines had a strict correlation to the knee effusion in OA. Many cytokines had a protective role and the others had to destroy the role of cartilage.
- ✓ The levels of four cytokines in both serum and SF in the same OA patients who had knee effusion.
- ✓ In the synovial fluid in OA patients, the IL-1β, IL-6, and IL-8 had a strict correlation to each other except IL-4 did not correlate with other cytokines. IL-4 in SF was significantly different between OA stage and pain level groups.

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References

1. Kluzek S, Sanchez-Santos MT, Leyland KM, Judge A, Spector TD, Hart D, et al. Painful knee but not hand osteoarthritis is an independent predictor of mortality over 23 years follow-up of a population-based cohort of middle-aged women. Ann Rheum Dis. 2016;75(10):1749-1756. doi: 10.1136/annrheumdis-2015-209026

2. Koneva ES, Lyadov KV, Shapovalenko TV, Zhukova EV, Polushkin VG. A personalized approach to the appointment of hardware techniques for restoring the walking stereotype in patients after total hip arthroplasty. Questions of balneology, physiotherapy and physical therapy. 2018;95(1):26-34. doi: 10.17116/kurort201895126-34

3. Disease GBD, Injury I, Prevalence C. Global, regional, and national incidence, prevalence, and years lived with disability for 354 diseases and injuries for 195 countries and territories, 1990–2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet. 2018;392:1789-1858.

4. 3. Ding SL, Pang ZY, Chen XM, Li Z, Liu XX, Zhai QL, et al. Urolithin a attenuates IL-1beta-induced inflammatory responses and cartilage degradation via inhibiting the MAPK/NF-kappaB signaling pathways in rat articular chondrocytes. J Inflamm (Lond). 2020;17(1):1-13. https://doi.org/10.1186/s12950-020-00242-8

5. Gu R, Shi Y, Huang W, Lao C, Zou Z, Pan S, Huang Z. Theobromine mitigates IL-1beta-induced oxidative stress, inflammatory response, and degradation of type II collagen in human chondrocytes. Int Immunopharmacol. 2020;82:106226. https://doi.org/10.1016/j.intimp.2020.106226

6. Jacques C, Gosset M, Berenbaum F, Gabay C. The role of IL-1 and IL-1Ra in joint inflammation and cartilage degradation. Vitam Horm. 2006;74:371-403.

7. Imamura M, Ezquerro F, Marcon Alfieri F, Vilas Boas L, Tozetto-Mendoza TR, Chen J, et al. Serum levels of proinflammatory cytokines in painful knee osteoarthritis and sensitization. Int J Inflam. 2015;2015:329792. https://doi.org/10.1155/2015/329792

8. Luo Z, Hu Z, Bian Y, Su W, Li X, Li S, et al. Scutellarin Attenuates the IL-1beta-Induced Inflammation in Mouse Chondrocytes and Prevents Osteoarthritic Progression. Front Pharmacol. 2020;11:107. https://doi.org/10.3389/fphar.2020.00107

9. Koh SM, Chan CK, Teo SH, Singh S, Merican A, Ng WM, et al. Elevated plasma and synovial fluid interleukin-8 and interleukin-18 may be associated with the pathogenesis of knee osteoarthritis. Knee. 2020;27(1):26-35. https://doi.org/10.1016/j.knee.2019.10.028

10. Ren G, Lutz I, Railton P, Wiley JP, McAllister J, Powell J, Krawetz R. Serum and synovial fluid cytokine profiling in hip osteoarthritis: distinct from knee osteoarthritis and correlated with pain. BMC Musculoskelet Disord. 2018;19(1):1-11. https://doi.org/10.1186/s12891-018-1955-4

11. Saetan N, Honsawek S, Tanavalee A, Yuktanandana P, Meknavin S, Ngarmukos S, et al. Relationship of plasma and synovial fluid vascular endothelial growth factor with radiographic severity in primary knee osteoarthritis. Int Orthop. 2014;38(5):1099-1104. https://doi.org/10.1007/s00264-013-2192-y

12. Fingleton C, Smart K, Moloney N, Fullen BM, Doody C. Pain sensitization in people with knee osteoarthritis: a systematic review and meta-analysis. Osteoarthritis Cartilage. 2015;23(7):1043-1056.

13. Hart DJ, Spector TD. The classification and assessment of osteoarthritis. Baillieres Clin Rheumatol. 1995;9(2):407-432.

14. Fernandes JC, Martel-Pelletier J, Pelletier JP. The role of cytokines in osteoarthritis pathophysiology. Biorheology. 2002;39(1-2):237-246.

15. Berenbaum F. Osteoarthritis as an inflammatory disease (osteoarthritis is not osteoarthrosis!). Osteoarthritis Cartilage. 2013;21(1):16-21. https://doi.org/10.1016/j.joca.2012.11.012

16. Palazzo C, Nguyen C, Lefevre-Colau MM, Rannou F, Poiraudeau S. Risk factors and burden of osteoarthritis. Ann Phys Rehabil Med. 2016;59(3):134-138. https://doi.org/10.1016/j.rehab.2016.01.006

17. de Rezende MU, de Campos GC. Is osteoarthritis a mechanical or inflammatory disease? Rev Bras Ortop. 2013;48(6):471-474. https://doi.org/10.1016/j.rboe.2013.12.002

18. Chow YY, Chin KY. The Role of Inflammation in the Pathogenesis of Osteoarthritis. Mediators Inflamm. 2020;2020:8293921. https://doi.org/10.1155/2020/8293921

19. Hunt MA, Charlton JM, Esculier JF. Osteoarthritis year in review 2019: mechanics. Osteoarthritis Cartilage. 2020;28(3):267-274. https://doi.org/10.1016/j.joca.2019.12.003

20. Yorimitsu M, Nishida K, Shimizu A, Doi H, Miyazawa S, Komiyama T, et al. Intra-articular injection of interleukin-4 decreases nitric oxide production by chondrocytes and ameliorates subsequent destruction of cartilage in instability-induced osteoarthritis in rat knee joints. Osteoarthritis Cartilage. 2008;16(7):764-771. https://doi.org/10.1016/j.joca.2007.11.006

21. Pelletier JP, DiBattista JA, Roughley P, McCollum R, Martel-Pelletier J. Cytokines and inflammation in cartilage degradation. Rheum Dis Clin North Am. 1993;19(3):545-568.

22. Liu CX, Gao G, Qin XQ, Deng CQ, Shen XJ. Correlation Analysis of C-terminal telopeptide of collagen type II and Interleukin-1beta for Early Diagnosis of Knee Osteoarthritis. Orthop Surg. 2020;12(1):286-294. https://doi.org/10.1111/os.12586

23. Attur MG, Patel IR, Patel RN, Abramson SB, Amin AR. Autocrine production of IL-1 beta by human osteoarthritis-affected cartilage and differential regulation of endogenous nitric oxide, IL-6, prostaglandin E2, and IL-8. Proc Assoc Am Physicians. 1998;110(1):65-72.

24. Wenham CY, Conaghan PG. New horizons in osteoarthritis. Age Ageing. 2013;42(3):272-278.

25. Penninx BW, Abbas H, Ambrosius W, Nicklas BJ, Davis C, Messier SP, Pahor M. Inflammatory markers and physical function among older adults with knee osteoarthritis. J Rheumatol. 2004;31(10):2027-2031.

26. Nees TA, Rosshirt N, Zhang JA, Reiner T, Sorbi R, Tripel E, et al. Synovial Cytokines Significantly Correlate with Osteoarthritis-Related Knee Pain and Disability: Inflammatory Mediators of Potential Clinical Relevance. J Clin Med. 2019;8(9):1343. https://doi.org/10.3390/jcm8091343

27. Ruan G, Xu J, Wang K, Zheng S, Wu J, Bian F, et al. Associations between serum IL-8 and knee symptoms, joint structures, and cartilage or bone biomarkers in patients with knee osteoarthritis. Clin Rheumatol. 2019;38(12):3609-3617. https://doi.org/10.1007/s10067-019-04718-8

28. Mabey T, Honsawek S, Tanavalee A, Yuktanandana P, Wilairatana V, Poovorawan Y. Plasma and synovial fluid inflammatory cytokine profiles in primary knee osteoarthritis. Biomarkers. 2016;21(7):639-644.

29. Goldring MB. Osteoarthritis and cartilage: the role of cytokines. Curr Rheumatol Rep. 2000;2(6):459-465.

30. Laavola M, Leppanen T, Hamalainen M, Vuolteenaho K, Moilanen T, Nieminen R, Moilanen E. IL-6 in Osteoarthritis: Effects of Pine Stilbenoids. Molecules. 2018;24(1): 109. https://doi.org/10.3390/molecules24010109

31. Jenei-Lanzl Z, Meurer A, Zaucke F. Interleukin-1beta signaling in osteoarthritis - chondrocytes in focus. Cell Signal. 2019;53:212-223.

32. McNulty AL, Rothfusz NE, Leddy HA, Guilak F. Synovial fluid concentrations and relative potency of interleukin-1 alpha and beta in cartilage and meniscus degradation. J Orthop Res. 2013;31(7):1039-1045. https://doi.org/10.1002/jor.22334

Tables

Table 1. The characteristic of the patients in the study

Variables	Patients number, n=35	
	Count	Percentage
Gender, n (%)		
Male	11	31.42
Female	24	68.58
The patients' age		
≤ 60	11	31.43
61 – 70	14	40.0
> 70	10	28.57
Disease duration		
≥ 60 months	7	20.0
< 60 months	28	80.0
Body Mass Index		
< 25	16	45.71
≥ 25	19	54.29
Visual analog scale score		
No pain	0	0
Mild	14	40
Moderate	21	80
Severy	0	0
Osteoarthritis grade based on radiography		
Grade 0	0	0
Grade 1	1	2.86
Grade 2	25 (71.43)	
Grade 3	9 (25.71)	
Grade 4	0	

Table 2. The level of cytokines in synovial fluid

		eIL-4	eIL-6	eIL-1β	eIL-8	p
Disease duration	≥ 60 months	6.11 ± 11.04	448.94 ± 724.25	1.64 ± 3.53	27.53 ± 36.05	p_{all} >0.05[a]
	< 60 months	5.77 ± 12.54	2021.91 ± 6347.08	11.41 ± 49.83	765.18 ± 2264.59	
VAS score groups	Mild	7.39 ± 13.86	182.05 ± 417.72	0.04 ± 0.01	5.96 ± 8.21	p_1= 0.035[b]
	Moderate	1.03 ± 0.39	2356.28 ± 6776.37	13.47 ± 53.39	886.13 ± 2415.61	p_{all} >0.05
BMI groups	< 25	6.19 ± 13.39	2111.17 ± 6826.50	12.67 ± 53.51	816.44 ± 2424.71	p_{all} >0.05
	≥ 25	5.82 ± 12.18	882.13 ± 924.23	1.17 ± 2.77	59.23 ± 54.69	
OA stages	Stage 2	7.34 ± 13.88	2230.69 ± 6800.31	13.28 ± 53.43	787.23 ± 2416.36	p_2 = 0.035[c]
	Stage 3	1.43 ± 0.51	544.86 ± 856.70	0.61 ± 1.33	291.66 ± 656.54	p_{all} >0.05

Note

VAS – visual analog scale;

eIL-4 : synovial fluid IL-4 ; eIL-6 : synovial fluid IL-6 ; eIL-8 : synovial fluid IL-8 ; eIL-1β : synovial fluid IL-1β ;

[a]p_{all} : p value for cytokines concentration between groups.

[b]p_1 : IL-4 in SF of mild VAS score group vs moderate VAS score group;

[c]p_2: IL-4 in SF of stage 2 vs stage 3;

Table 3: The correlation between OA stage, VAS score and concentration of four interleukins in serum and synovial fluid

	VAS	sIL-4	eIL-4	sIL-6	eIL-6	sIL-1β	eIL-1β	sIL-8	eIL-8

	core								
OA stage	0.114	-0.115	-0.171	-0.082	-0.097	-0.109	-0.097	-0.068	-0.076
VAS score		0.211	0.168	0.213	0.319	0.245	0.287	0.209	0.332
sIL-4			-0.087	**0.951**[†]	0.077	**0.791**[†]	-0.053	**0.949**[†]	-0.048
eIL-4				-0.067	-0.005	0.000	-0.034	-0.072	0.014
sIl-6					0.088	**0.833**[†]	-0.038	**0.999**[†]	-0.026
eIL-6						0.039	**0.966**[†]	0.083	**0.732**[†]
sIL-1β							-0.062	**0.833**[†]	-0.061
eIL-1β								-0.041	**0.851**[†]
sIL-8									-0.031

Notes:

OA – osteoarthritis;

VAS - visual analog scale;

[†]Correlation was significant with p < 0.001 (2-tailed). [††] Correlation was significant with p<0.05 (2-tailed).

(OA : osteoarthritis ; sIL-4: serum IL-4; sIL-6: serum IL-6 ; sIL-8 : serum IL-8 ; sIL-1β : serum IL-1β ; eIL-4 : synovial fluid IL-4 ; eIL-6 : synovial fluid IL-6 ; eIL-8 : synovial fluid IL-8 ; eIL-1β : synovial fluid IL-1 β).

The interleukin levels did not correlate to disease stage and VAS score. In synovial fluid, the concentration of IL-1β, IL-8 and IL-6 had high correlation to each others. In the serum, all four interleukin level correlated highly to each other.

Figures

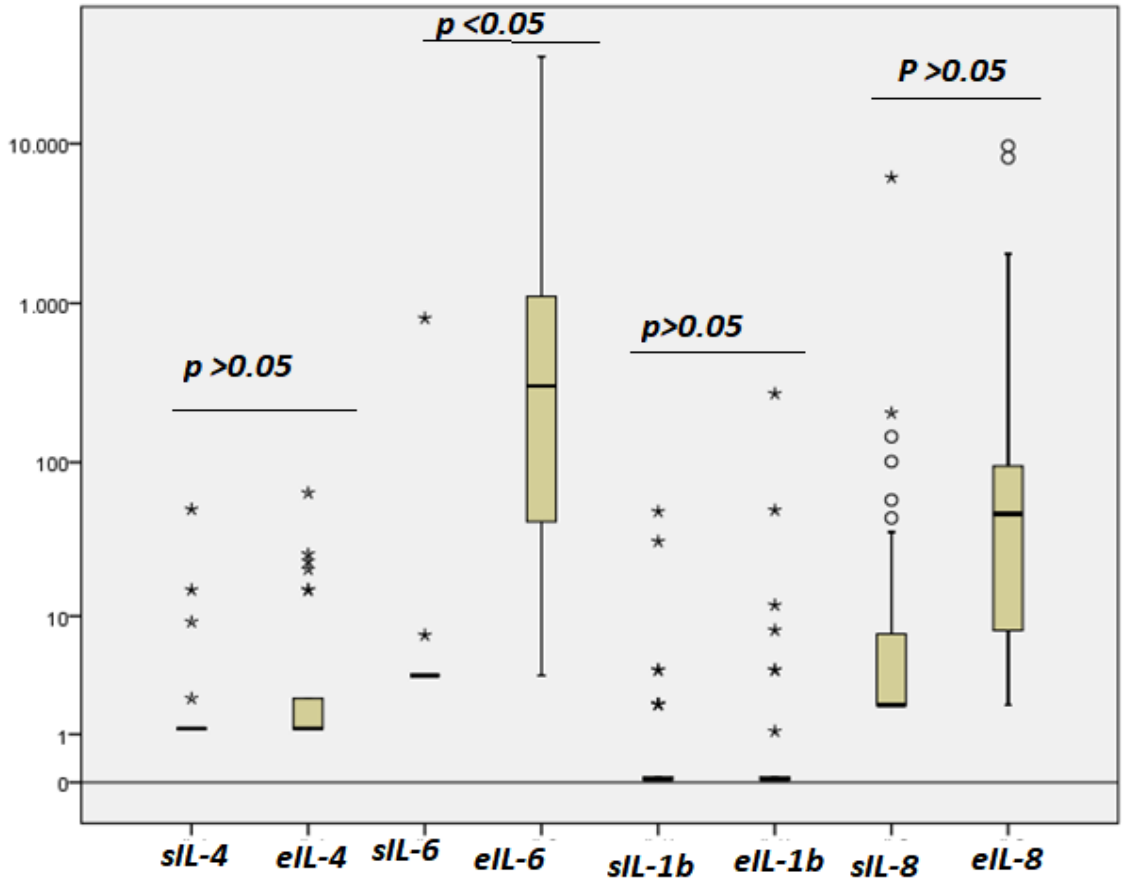

Figure 1. The level of IL-4, IL-6, IL-1β, IL-8 in serum and synovial fluid

The level of IL-6 in serum and synovial fluid had significant difference ($p< 0.05$). The level of the others in serum and synovial fluid were not different significantly.

(sIL-4: serum IL-4; sIL-6: serum IL-6 ; sIL-8 : serum IL-8 ; sIL-1β : serum IL-1β ; eIL-4 : synovial fluid IL-4 ; eIL-6 : synovial fluid IL-6 ; eIL-8 : synovial fluid IL-8 ; eIL-1β : synovial fluid IL-1 β).