Markedly elevated serum IL-6 levels predict relapse within six months of treatment initiation in Still's disease

Sirs,

Still's disease (SD) is a systemic autoinflammatory disease. Pathogen- or damage-associated molecular patterns activate the NACHT, LRR, and PYD domains of the protein 3 inflammasome, leading to the overproduction of inflammatory cytokines such as interleukin (IL)-1β, IL-6, IL-18, and tumour necrosis factor-α (1). The serum levels of IL-6 and IL-18 correlate with disease activity (1). Correlations between cytokines and clinical subtypes have been reported: the IL-6-dominant pattern is associated with arthritis (2), and the IL-18-dominant pattern is associate with systemic symptoms and pleuritis (1, 3). However, the impact of these cytokines, especially serum IL-6, on the prognosis in patients with SD has not been thoroughly investigated. In this study, the effects serum levels of IL-6 and IL-18 of baseline on the prognosis of these patients were investigated.

Eleven patients with SD, diagnosed using Yamaguchi's criteria (4), who exhibited high disease activity treated at the Kobe City Medical Center General Hospital from June 2022 to April 2023 were included in this study. The study was conducted in accordance with the Declaration of Helsinki and approved by the ethics committee of our institution (approval no.: zn231212).

The patients were categorised into two groups: group 1 with markedly elevated serum IL-6 levels and group 2 with moderately elevated levels. High disease activity was defined as a modified Pouchot score ≥4 (5) at the time of onset or relapse. Relapse was characterised by specific clinical symptoms, laboratory abnormalities and the necessity for increased doses of immunosuppressants or glucocorticoids (GCs). The serum levels of IL-6 and IL-18 were measured prospectively, and the patients' medical records, including clinical characteristics, blood tests (including cytokines), treatment and clinical progression, were reviewed. The serum IL-6 and IL-18 levels were assessed using the chemiluminescent enzyme immunoassay and enzyme immunoassay methods, respectively, at LSI Medience Corporation, Japan.

Hierarchical cluster analysis (Ward's method) was used based on the serum IL-6 and IL-18 levels of the 11 patients with high disease activity (Fig. 1A). The patients were classified into two groups. All three patients in group 1 exhibited markedly elevated serum levels of both IL-6 and IL-18 (Fig. 1B). Nine patients were undergoing initial treatment, two were experiencing relapse and one patient was administered tocilizumab (Table I). Macrophage activation syndrome was not observed in any patients in this study. All patients were administered high-dose GCs (prednisolone 1-2 mg/kg/day with or without a methylprednisolone pulse). The clinical and demographic features at onset or relapse within six months of treatment initiation were not significantly different between the groups, except for the serum IL-6 levels (group 1 median: 628 pg/mL; group 2 median: 79 pg/mL; p=0.0189) and relapse within six months (all three patients in group 1 and no patients in group 2; p=0.0061). The three patients in group 1 who experienced relapse were resistant to high-dose GC, cyclosporin A, and intravenous tocilizumab (8 mg/kg weekly). The serum IL-18 levels tended to be higher in group 1 (median: 198,000 pg/mL) than in group 2 (median: 113,650 pg/mL); however, this difference was not significant.

Serum IL-6 levels >600 pg/mL prior to the initiation of high-dose GC in patients with high disease activity may predict relapse within six months of treatment. In patients with rheumatoid arthritis, high serum IL-6 levels (mean: 95.33±128.7 pg/ml) were associated with poor response to tocilizumab (6), which is consistent with the results of the current study regarding patients with SD. In this study, the three patients with high disease activity and extremely high serum IL-6 levels were resistant to intensive treatment, including tocilizumab. These patients may require anti-IL-1 therapy or a high dose of tocilizumab.

Serum IL-18 levels have been used as an indicator of disease activity and severity in patients with SD (1) and have been reported as predictors of poor treatment efficacy at week 24 (7). However, the results of the current study suggest that IL-6 predicts the treatment response more accurately than IL-18. This finding was observed even when patients were treated with tocilizumab. IL-6 typically increases when tocilizumab is used; IL-6 levels of 89.7 pg/mL have been reported 42 days after treatment (baseline level: 58.4 pg/mL) in patients with rheumatoid arthritis (8). The results of the current study suggest that measuring the serum IL-6 level is useful even when the patients are treated with tocilizumab or an IL-6 receptor antagonist.

Assessing the serum IL-18 and IL-6 levels prior to treatment allows for the prediction of treatment response and relapse within six months of treatment, providing valuable information for patient management. To the best of our knowledge, this is the first study to demonstrate that very high baseline serum IL-6 levels may predict relapse within six months of treatment in patients with SD.

In summary, markedly elevated baseline serum IL-6 levels at disease or relapse onset may predict subsequent relapse within six months despite aggressive treatment and resistance to high doses of glucocorticoids, cyclosporin A, and tocilizumab. Further validation with a larger cohort of patients is required.

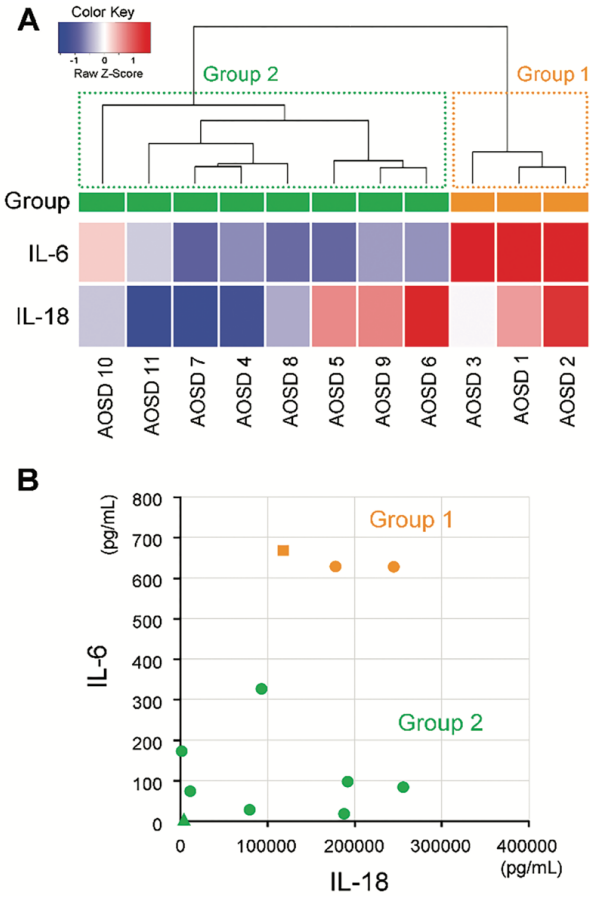

Fig. 1. Patients with high disease activity had very high or high levels of serum IL-6 and IL-18.
A: The hierarchical cluster analysis of the serum IL-6 and IL-18 levels of 11 patients with high disease activity, resulting in their classification into two groups is shown.
B: A scatter plot depicting the serum IL-6 and IL-18 levels in the same 11 patients with high disease activity is shown. Group 1 has higher serum IL-6 levels than group 2. The square plot indicates relapse during treatment with tocilizumab in high disease activity periods. The triangle plot indicates relapse during treatment without tocilizumab in high disease activity periods.

Table I. Patient characteristics.

	Group 1 (n=3)	Group 2 (n=8)	*p*-value
Female, n (%)	2 (67)	8 (100)	0.2727
Age	54 (51, 65)	61 (44, 73)	1.0000
Duration, years	0 (0, 2)	0 (0, 0)	0.4491
Symptoms at high disease activity			
Fever, n (%)	3 (100)	8 (100)	0.0000
Arthralgia, n (%)	3 (100)	7 (88)	1.0000
Rash, n (%)	2 (67)	7 (88)	0.4909
Leukocytosis, n (5)	3 (100)	8 (100)	0.000
Sore throat, n (%)	3 (100)	4 (50)	0.2364
Lymphadenopathy, n (%)	2 (67)	4 (50)	1.0000
Liver dysfunction, n (%)	2 (67)	7 (88)	0.4909
Splenomegaly, n (%)	2 (67)	3 (25)	0.5455
Serositis, n (%)	1 (33)	1 (13)	0.4909
Laboratory data at high disease activity			
WBC, /μL	20,400 (16250, 29700)	21,000 (16625, 22475)	0.9187
Neutrophil, %	92 (90.3, 93)	91 (87.5, 92.8)	0.6082
Haemoglobin, g/dL	10 (9.7, 10.6)	11 (10.9, 11.6)	0.3583
Platelet, $\times 10^4/\mu L$	30 (18, 33.3)	28 (21.0, 38.8)	0.7595
AST, U/L	50 (44, 235)	67 (43, 105)	1.0000
ALT, U/L	51 (33, 138)	64 (29, 92)	1.0000
LD, U/L	740 (590, 1161)	450 (421, 541)	0.3583
sIL-2R, U/mL	3855 (2853, 4016)	2013 (1248, 2906)	0.2616
Ferritin, ng/mL	37728 (24647, 39623)	8272 (1331, 18563)	0.2616
CRP, mg/dL	38 (29.3, 38.5)	25 (16.8, 27.6)	0.3583
IL-6, pg/mL	628 (627.5, 647.5)	79 (25.2, 93.6)	**0.0189***
IL-18, pg/mL	198000 (158000, 221500)	113650 (9308, 206500)	0.4750
Relapse at high disease activity, n (%)	1 (33)	1 (13)	0.4449
Use of tocilizumab in relapsed patient, n (%)	1 (33)	0 (0)	
Treatment within six months			
Prednisolone at six months or relapse, mg/day	7 (6, 16)	7 (5, 7.3)	0.6731
Maximum prednisolone, mg/day	80 (63, 100)	55 (49, 61)	0.3041
Cyclosporin A, n (%)	3 (100)	4 (50)	0.2364
Methotrexate, n (%)	1 (33)	2 (25)	1.0000
Tocilizumab, n (%)	3 (100)	5 (63)	0.4909
Relapse within six months, n (%)	3 (100)	0 (0)	**0.0061***

Continuous variables are presented as the median and the first and third quartiles. These were compared using the Mann-Whitney U-test. The Fisher's exact test was used to analyse categorical variables. Statistical significance was set at $p<0.05$.

Normal ranges: WBC: 3900-9800 /μL; neutrophil: 26-71%; platelets: 13-37×10⁴/μL; haemoglobin: 11.1-15.1 g/dL; AST: 8-40 U/L; ALT: 8-40 U/L; LD: 122-222 U/L; sIL-2R: 122-496 U/mL; ferritin: 2.6-129.4 ng/mL; CRP: 0.0-0.5 mg/dL.

GC: glucocorticoids; WBC: white blood cell; Hb: haemoglobin; AST: aspartate aminotransferase; ALT: alanine aminotransferase; LD: lactate dehydrogenase; sIL-2R: soluble interleukin-2 receptor.

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References

1. FEIST E, MITROVIC S, FAUTREL B: Mechanisms, biomarkers and targets for adult-onset Still's disease. *Nat Rev Rheumatol* 2018; 14(10): 603-18. https://doi.org/10.1038/s41584-018-0081-x
2. INOUE N, SHIMIZU M, TSUNODA S, KAWANO M, MATSUMURA M, YACHIE A: Cytokine profile in adult-onset Still's disease: Comparison with systemic juvenile idiopathic arthritis. *Clin Immunol* 2016; 169: 8-13. https://doi.org/10.1016/j.clim.2016.05.010
3. NAM SW, KANG S, LEE JH, YOO DH: Different features of interleukin-37 and interleukin-18 as disase activity markers of adult-onset Still's disease. *J Clin Med* 2021; 10(5): 910. https://doi.org/10.3390/jcm10050910
4. YAMAGUCHI M, OHTA A, TSUNEMATSU T *et al*.: Preliminary criteria for classification of adult Still's disease. *J Rheumatol* 1992; 19(3): 424-30.
5. RAU M, SCHILLER M, KRIENKE S, HEYDER P, LORENZ H, BLANK N: Clinical manifestations but not cytokine profiles differentiate adult-onset Still's disease and sepsis. *J Rheumatol* 2010; 37(11): 2369-76. https://doi.org/10.3899/jrheum.100247
6. SHIMAMOTO K, ITO T, OZAKI Y *et al*.: Serum interleukin 6 before and after therapy with tocilizumab is a principal biomarker in patients with rheumatoid arthritis. *J Rheumatol* 2013; 40(7): 1074-81. https://doi.org/10.3899/jrheum.121389
7. TANG KT, HSIEH CW, CHEN HH *et al*.: The effectiveness of tocilizumab in treating refractory adult-onset Still's disease with dichotomous phenotypes: IL-18 is a potential predictor of therapeutic response. *Clin Rheumatol* 2022; 41(2): 557-66. https://doi.org/10.1007/s10067-021-05921-2
8. NISHIMOTO N, TERAO K, MIMA T, NAKAHARA H, TAKAGI N, KAKEHI T: Mechanisms and pathologic significances in increase in serum interleukin-6 (IL-6) and soluble IL-6 receptor after administration of an anti-IL-6 receptor antibody, tocilizumab, in patients with rheumatoid arthritis and Castleman disease. *Blood* 2008; 112(10): 3959-64. https://doi.org/10.1182/blood-2008-05-155846