

# Interleukin 18: A Biomarker for Differential Diagnosis Between Adult-onset Still's Disease and Sepsis

Roberta Priori, Serena Colafrancesco, Cristiano Alessandri, Antonina Minniti, Carlo Perricone, Giancarlo Iaiani, Donatella Palazzo, and Guido Valesini

**ABSTRACT. Objective.** The differential diagnosis between rheumatic diseases and infectious conditions is a great challenge in clinical practice. Adult-onset Still's disease (AOSD) is a rare systemic inflammatory syndrome that shares several clinical and laboratory variables with sepsis. Interleukin (IL)-18 is overexpressed in AOSD, suggesting a possible role as a disease biomarker. The aim of our study was to detect IL-18 serum levels in a cohort of patients with AOSD and sepsis and to address its possible role as a biomarker for differential diagnosis.

**Methods.** A group of unselected patients with AOSD diagnosed according to the Yamaguchi criteria and consecutive patients with sepsis diagnosed according to the American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference criteria were enrolled. The clinical and laboratory data were collected. In the AOSD group, disease activity was assessed by Pouchot's and Rau's criteria. IL-18 serum levels were detected by ELISA.

**Results.** Thirty-nine patients with AOSD and 18 patients with sepsis were enrolled. Two out of 18 patients with sepsis (11.1%) also fulfilled the Yamaguchi criteria. A significant difference was found in IL-18 serum levels between patients with active and inactive disease ( $p < 0.001$ ), and it positively correlated with disease activity ( $p = 0.0003$ ), ferritin serum level ( $p = 0.016$ ), and erythrocyte sedimentation rate ( $p = 0.041$ ). IL-18 was significantly increased in patients with AOSD when compared with sepsis ( $p = 0.014$ ). For a cutoff of 148.9 pg/ml, this test had a specificity of 78.3% and a sensitivity of 88.6%.

**Conclusion.** We have demonstrated that IL-18 can be a biomarker for differential diagnosis between AOSD and sepsis. (J Rheumatol First Release May 1 2014; doi:10.3899/jrheum.130575)

#### Key Indexing Terms:

ADULT-ONSET STILL'S DISEASE      SEPSIS      INTERLEUKIN 18      FERRITIN

The differential diagnosis between rheumatic diseases and infectious conditions is frequently a great challenge in clinical practice. Adult-onset Still's disease (AOSD) is a rare systemic inflammatory syndrome characterized by the presence of a triad of symptoms composed of high daily spiking fever, evanescent salmon-colored rash, and arthritis or arthralgia<sup>1</sup>. Different diagnostic criteria for AOSD have been proposed, but the most commonly used are those published by Yamaguchi, *et al* in 1992<sup>2</sup>. These criteria

underline that AOSD represents a diagnosis of exclusion in which other conditions such as rheumatic diseases, malignancies, and infections need to be ruled out. Indeed, the clinical manifestations in AOSD are often similar to some infectious conditions such as sepsis. Among the shared clinical features of these 2 entities, fever is a key sign, often associated with a wide spectrum of clinical manifestations such as arthralgia, myalgia, hepatosplenomegaly, lymphadenopathy, and skin eruptions. The similarities between AOSD and sepsis encompass also several laboratory aspects such as a marked neutrophilic leukocytosis, increased liver enzymes, or hyperferritinemia. A new entity called hyperferritinemic syndrome<sup>3</sup>, in which AOSD and sepsis are included, has been suggested recently. In this condition, a possible pathogenic role of ferritin leading to the development of a cytokine storm has been hypothesized.

Therefore, biomarkers are needed with high sensitivity and specificity, useful not only to dynamically measure AOSD disease severity and to predict prognosis, but also to distinguish this from other inflammatory conditions. Efforts have been made to unveil the immunologic and molecular basis of these conditions, and several putative mediators have been identified so far. Among them, interleukin 18

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(IL-18), an immunoregulatory and inflammatory cytokine, has attracted interest as a potential therapeutic target in autoimmune/inflammatory disorders<sup>4</sup>. We confirmed the presence of high serum levels of IL-18 in AOSD and demonstrated its possible role as a disease biomarker<sup>5</sup>. This cytokine seems to be overexpressed also in patients with sepsis, where the inflammatory process is driven by a cytokine storm<sup>6</sup>. Thus, the aim of our study was to detect IL-18 serum level in a cohort of patients with AOSD and sepsis and to address its possible role as a biomarker for differential diagnosis.

## MATERIALS AND METHODS

A group of unselected patients with AOSD diagnosed according to the Yamaguchi criteria<sup>2</sup> and followed at the Rheumatology Unit of Sapienza University of Rome and consecutive patients with sepsis diagnosed according to the American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference criteria<sup>7</sup> and hospitalized at the Infectious Disease Unit of the same hospital were enrolled in our study. Patients were not matched for age and sex owing to the rarity of the disease. For this reason we evaluated all the possible serum samples collected from patients with AOSD during the last 10 years in our unit. The clinical and laboratory data [erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), white blood cell count (WBC), and ferritin] were collected in both groups. Patients of the sepsis group were also screened to verify whether they fulfilled the Yamaguchi criteria. In the AOSD group, disease activity was assessed either by the criteria proposed by Pouchot in 1991<sup>8</sup> or by those modified by Rau in 2010<sup>9</sup>. In both sets of criteria, the total score ranges from 0 to 12 and is calculated through the addition of points assigned to each symptom. The Pouchot's score uses the following items: fever, evanescent rash, pleurisy, pneumonia, pericarditis, hepatomegaly or abnormal liver function tests, splenomegaly, lymphadenopathy, WBC > 15,000/mm<sup>3</sup>, sore throat, myalgias, and abdominal pain. In the score proposed by Rau, "splenomegaly" and "abdominal pain" were changed to "ferritin serum levels  $\geq$  3000  $\alpha$ g/l", and "arthritis". Patients with a Pouchot's or Rau's score  $\geq$  4 were considered to have active disease<sup>9</sup>. Blood samples were collected from patients with active and inactive AOSD and from patients with sepsis during the febrile attack (first 48 h of hospitalization). IL-18 serum level was detected by means of ELISA (Immuno Pharmacology Research) as previously described<sup>5</sup>. All subjects provided their informed consent.

The values were expressed as mean  $\pm$  SD. For the statistical analysis, the Mann-Whitney U test and Spearman's rank correlation test were used. Two-tailed p values < 0.05 were considered significant. Area under the receiver-operating characteristic curve (ROC-AUC) analysis was used to evaluate the diagnostic utility of the IL-18 serum level. The evaluation of the laboratory features between patients with active and inactive disease was performed using Rau's criteria.

## RESULTS

Thirty-nine patients with AOSD (F = 25, 64.1%; M = 14, 35.9%; mean age  $\pm$  SD = 35  $\pm$  14 yrs, mean age at onset  $\pm$  SD = 32  $\pm$  12 yrs) and 18 patients with sepsis (F = 8, 44.4%; M = 10, 55.6%; mean age  $\pm$  SD = 54  $\pm$  17.4 yrs) were enrolled. Seventeen out of 39 (43.5%) and 23 out of 39 (58.9%) patients with AOSD had active disease when the serum was taken, according to Pouchot's criteria and Rau's criteria, respectively. Two of the 18 patients with sepsis (11.1%) also fulfilled the Yamaguchi criteria. Main clinical

features from patients with AOSD and sepsis are reported in Table 1. A significant difference in ferritin serum level was found between patients with active and inactive AOSD (active disease mean  $\pm$  SD = 6462.4  $\pm$  8427.7, inactive disease mean  $\pm$  SD = 416.7  $\pm$  466.8; p = 0.039). IL-18 mean value in sera of the whole cohort of patients with AOSD was 1043.9  $\pm$  1831.4 pg/ml. A significant difference was found in IL-18 serum level between patients with active disease and patients with inactive disease (1716.8  $\pm$  2182.6 vs 118.7  $\pm$  159.2 pg/ml, p < 0.001; Figure 1). IL-18 serum level positively correlated with disease activity (p = 0.0003; Figure 2A), ferritin serum level (p = 0.016; Figure 2B), and ESR (p = 0.041; Figure 2C). In patients with sepsis, mean IL-18 serum level was 91.2  $\pm$  73.3 pg/ml, thus significantly lower when compared with the whole AOSD cohort (p = 0.014; Figure 1) as well as only with the patients with inactive disease (p < 0.0001; Figure 1). The area under the ROC-AUC for detection of AOSD by reference to the level of serum IL-18 was 0.864 (Figure 3). For a cutoff of IL-18 of 148.9 pg/ml, this test had a specificity of 78.3% and a sensitivity of 88.6%; likelihood ratio was 2.89. Concerning other laboratory features, there was no significant difference in ESR (p = 0.769), CRP (p = 0.62), WBC (p = 0.14), and ferritin serum level (p = 0.27) between AOSD and sepsis (Table 2). Moreover, in patients with sepsis, IL-18 level did not correlate with any of the serological variables: ESR (p = 0.79), CRP (p = 0.46), WBC (p = 0.98), and ferritin (p = 0.10).

## DISCUSSION

To our knowledge, we demonstrated for the first time that IL-18 can be a biomarker for differential diagnosis between AOSD and sepsis. Prompt and correct diagnosis of these life-threatening conditions may allow proper treatment and lifesaving action<sup>10</sup>. Sometimes a clinical and serological overlap between AOSD and sepsis can be observed. Indeed, about 11% of our patients with sepsis also fulfilled the Yamaguchi criteria, and the diagnosis could have been puzzling. In our opinion, this evidence supports the idea that, at least in a small percentage of cases, clinical aspects

Table 1. Clinical features in 39 patients with adult-onset Still's disease (AOSD) and 18 with sepsis.

Signs and Symptoms	AOSD (% of patients)	Sepsis (% of patients)
Fever	100	100
Rash	76.9	9
Arthritis	64.1	9
Sore throat	64.1	9
Lymphadenopathy	58.9	27.2
Hepatomegaly/increased liver enzymes	48.7	45.4
Myalgia	38.4	18.1
Splenomegaly	15.3	36.3
Pericarditis	10.2	0
Pleurisy	7.6	0

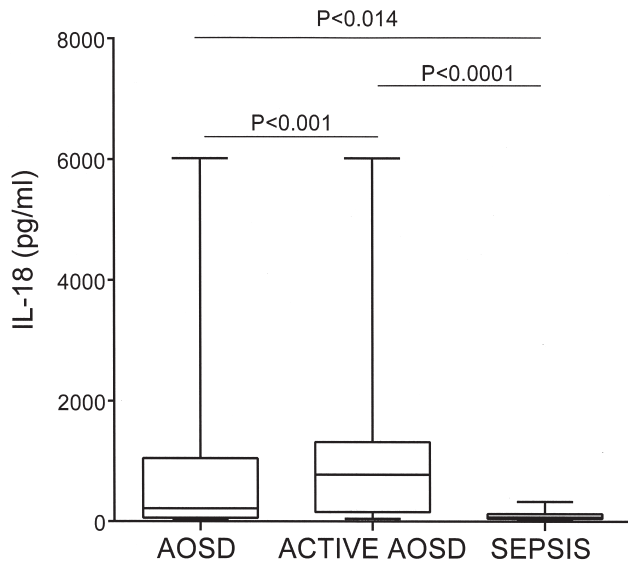


Figure 1. Comparison between IL-18 serum levels in the whole cohort of patients with AOSD (n = 39), patients with active disease (n = 17), and sepsis (n = 18). AOSD: adult-onset Still's disease; IL: interleukin.

and routine laboratory tests may not always allow the physician to achieve the correct diagnosis. Moreover, it is possible that during sepsis the microbiological tests such as blood cultures are negative. For this reason, the use of specific biomarkers able to characterize each condition is becoming more and more essential. Biomarkers have an important role not only in the diagnostic process but also in the assessment of disease prognosis, in the choice of therapeutic approach, in monitoring response to therapy, and in evaluation of remission. In sepsis, several biomarkers have been proposed including cytokines, cell markers, receptors, coagulation constituent, vascular endothelial damage molecule, vasodilation-related molecules, acute-phase reactants, and organ-damaging molecules<sup>11</sup>. Procalcitonin (PCT) has been proposed as a more specific and better prognostic marker in infectious conditions<sup>12</sup>, although its value has also been challenged. In a recent work, Scirè, *et al*<sup>13</sup> demonstrated that, although in different rheumatic diseases PCT can discriminate between infectious and noninfectious etiology of fever, it does not seem useful for AOSD. In patients with AOSD and fever, an overexpression of PCT can be found even in the absence of infection<sup>12</sup>.

For this reason, to identify a possible biomarker in patients with AOSD, several studies have been undertaken. Kawashima, *et al*<sup>14</sup> were the first to demonstrate the markedly increased expression of IL-18 in systemic circulation of patients with AOSD. Later on, Kawaguchi, *et al*<sup>15</sup>, proposed IL-18 not only as a diagnostic marker for AOSD but also as an indicator of disease severity. IL-18 is considered a member of the IL-1 superfamily because of its structural homology and biological effects. IL-1 seems to have a pathogenic role in several autoinflammatory condi-

tions and its production is driven by the “inflammasome” activation. Indeed, the inflammasome induction can be mediated by different innate immune receptors (Toll-like receptors or NOD-like receptors) previously activated by external pathogens (pathogen-associated molecular patterns) or by endogenous molecules (damage-associated molecular patterns). Such structures mediate the innate immune system response, promoting the production of inflammatory caspases and activating proinflammatory cytokines such as IL-1 $\beta$  and IL-18<sup>16</sup>. Because of the promising results obtained by the use of anti-IL-1 agents in AOSD as well as in other autoinflammatory conditions, modulation of these pathways is now the main therapeutic target. Following its production, IL-18 interacts with a complex receptor (IL-18R), which is widely expressed on cells implicated in innate and acquired immunity<sup>4</sup>. IL-18 was initially identified as a major inducer of interferon (IFN)- $\gamma$ <sup>17</sup> and is a critical regulator of Th1 cell maturation. Otherwise, depending on the context of stimulation, IL-18 may promote either Th1 or Th2 response<sup>18</sup>.

It has been demonstrated that not only classic antigen-presenting cells, but also nonimmune cells such as synovial fibroblasts, intestinal epithelial cells, and keratinocytes, contribute in a proinflammatory manner as IL-18-producing cells. Thus, IL-18 provides a critical link between innate and acquired immune responses with the involvement of both nonprofessional and professional immune cell lineages<sup>4</sup>. This is why an increasing interest in the role of this cytokine has been emerged in connection with different kinds of autoimmune/inflammatory disorders such as AOSD.

In previous studies, we have shown that IL-18 is overexpressed in sera<sup>5</sup> as well as in lymph nodes<sup>19</sup> and liver<sup>20</sup> of patients with AOSD. Otherwise, it has been suggested that IL-18 plays an important role in the pathophysiology of sepsis<sup>6,21,22,23</sup>. In infections, this cytokine has a key function because of its ability to regulate innate and acquired immune response. Further, it has been demonstrated that it participates in the defense against intracellular bacteria including *Listeria*, *Shigella*, *Salmonella*, and *Mycobacterium tuberculosis*<sup>24</sup>. A study by Marshall, *et al*<sup>25</sup> demonstrated that stimulation of blood monocytes with *Staphylococcus aureus* causes a superinduction of IL-18 compared with lipopolysaccharide. Thereafter, a comparison of plasma IL-18 concentrations between patients with gram-positive and gram-negative sepsis showed significantly higher levels of this cytokine in the group with gram-positive infection<sup>26</sup>.

Even though the exact downstream signaling mechanism of inflammation in course of sepsis has not been fully uncovered, it appears that a selected group of inflammatory mediators such as tumor necrosis factor- $\alpha$  (TNF- $\alpha$ ), IL-1 $\beta$ , IL-6, IL-7, IL-8, IL-10, IL-13, and IFN- $\gamma$  could be involved. Those mediators proved to be associated with the severity and evolution of organ dysfunction in sepsis<sup>27</sup>. Within this

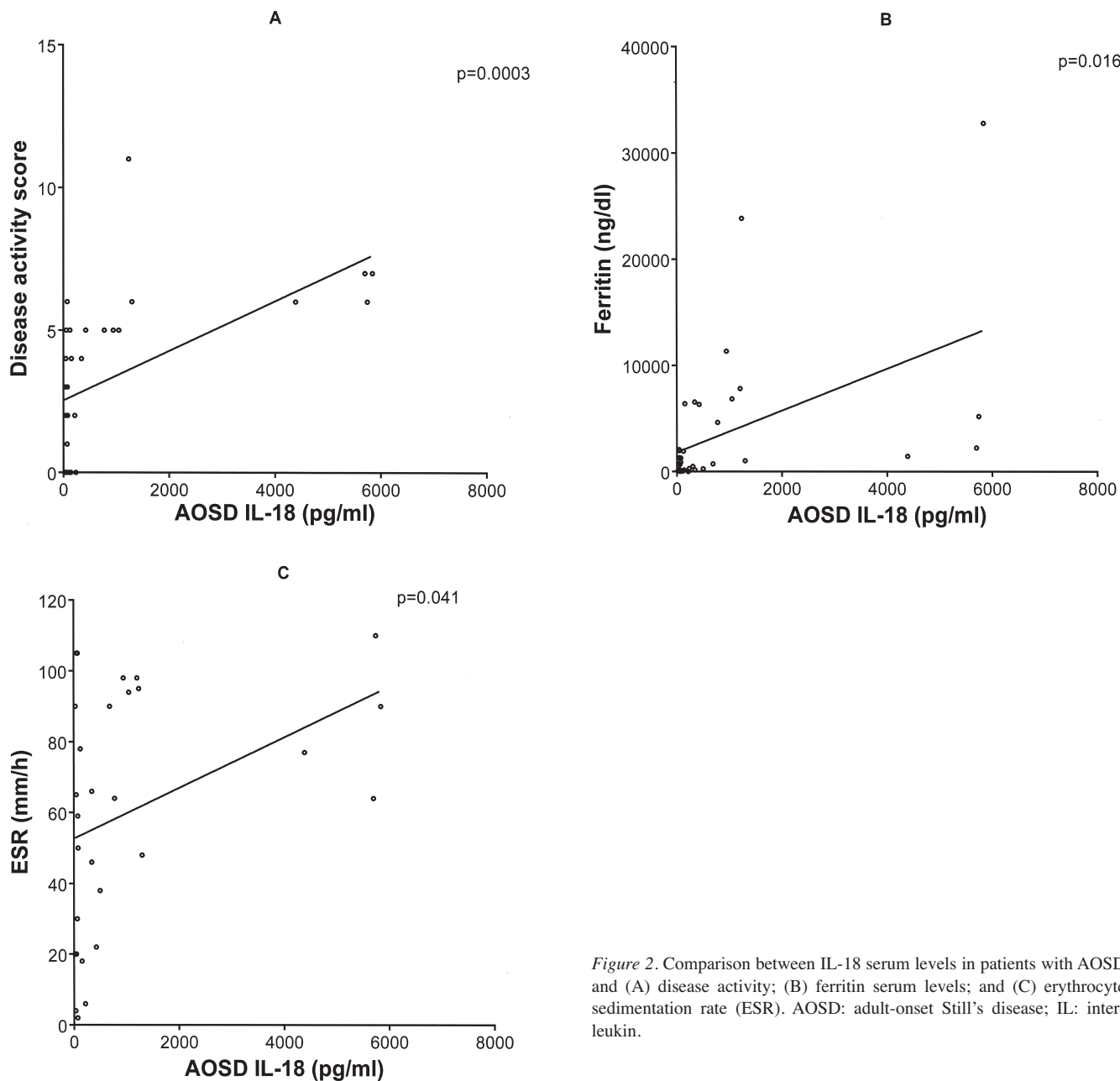


Figure 2. Comparison between IL-18 serum levels in patients with AOSD and (A) disease activity; (B) ferritin serum levels; and (C) erythrocyte sedimentation rate (ESR). AOSD: adult-onset Still's disease; IL: interleukin.

group of cytokines, IL-18 seems to make the major contribution as central initiator and regulator of the inflammatory cascade<sup>6</sup>. Nonetheless, Rau, *et al*<sup>9</sup> compared a panel of serum inflammatory molecules (including IL-1 $\beta$ , IL-6, IL-8, IL-10, IL-12, IFN- $\gamma$ , TNF, and calprotectin, as well as IL-18) without finding any significant difference between patients with sepsis and those with AOSD. The major limitation of that study was the low number of patients in the study cohort (18 patients with AOSD and 14 sepsis). Further, as the authors underlined, the observed broad range of IL-18 levels in all groups of patients may have biased the statistical analysis.

Interestingly, our results may integrate these findings. IL-18 serum level was significantly increased in the AOSD group when compared with sepsis, considering the whole cohort of patients and only the patients with active disease. Apparently, in our study, patients with sepsis presented IL-18 serum levels lower than previously reported<sup>21</sup>. We cannot exclude that those levels may depend on the duration of septic disease and treatment. Indeed, although all the serum samples were taken within 48 h of hospitalization, some patients had already been treated with antibiotics at home. Moreover, such a finding is not surprising in light of the known variability described for IL-18 in the course of



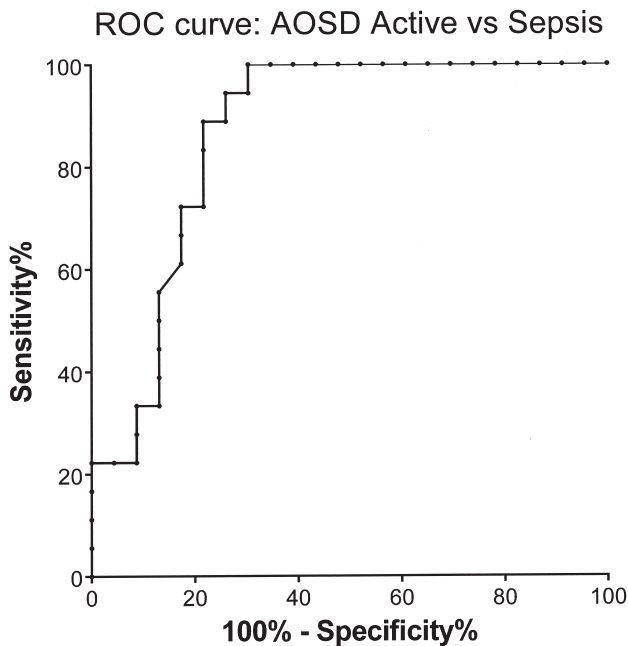


Figure 3. Area under the receiver-operating characteristic curve (ROC) for detection of AOSD by reference to the level of serum IL-18. AOSD: adult-onset Still's disease.

Table 2. Laboratory features in patients with AOSD and sepsis. Except for p value, data are mean ( $\pm$  SD).

Features	AOSD	Sepsis	p
ESR, mm/h	60.4 (34.2)	61.3 (33.9)	0.769
CRP, mg/dl	73.2 (84.4)	116.4 (98.2)	0.62
WBC, cells/ $\mu$ l	15,822 (8038)	11,192.5 (7120.5)	0.14
Ferritin, ng/ml	3540.3 (6701.9)	1720 (3882)	0.27
IL-18, pg/ml	1043.9 (1831.4)	91.2 (73.3)	0.014

AOSD: adult-onset Still's disease; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; WBC: white blood cells; IL: interleukin.

sepsis<sup>28</sup>. A wide range of serum values has been documented<sup>29,30</sup>, suggesting how such variability may be ascribed to different aspects, including the methodology used for cytokine detection<sup>31</sup> or the different infectious agents responsible for sepsis<sup>6</sup>. Rau and colleagues<sup>9</sup> agreed, and their study did not show the data on IL-18 because of a too-broad range of values. This may lead us to hypothesize very low levels of IL-18 in the patients with sepsis in their study too. Further, in our study serum IL-18 detection was performed using an ELISA test, while in Rau's a multiplex analysis was undertaken. As reported by Leng, *et al*<sup>31</sup>, experience with multiplex arrays remains limited, particularly in the context of human aging studies, and although good correlations between ELISA and multiplex have been reported, careful side-by-side comparisons are rare. Moreover, because there are no World Health Organiza-

tion-accepted standards for normal age-adjusted cytokine levels, comparing results obtained using single ELISA kits from different manufacturers or comparing singleplex to multiplex results can be problematic<sup>31</sup>.

Yet another consideration has to be taken in account. In 2001, Novick, *et al*<sup>30</sup> measured serum levels of both IL-18 and IL-18 binding protein (IL-18BP) in a large cohort of patients with sepsis. The authors found that total IL-18 and its binding protein were elevated, and most of the IL-18 was bound to IL-18BP. The authors concluded that the binding protein considerably inhibits the free circulating form of IL-18, thus giving rise to lower detectable levels.

In our study, IL-18 was the only serological variable that distinguished the 2 groups. No significant differences between AOSD and sepsis were found in ESR, CRP, WBC, and ferritin levels. Moreover, IL-18 serum level correlated with other serological markers of inflammation only in patients with AOSD. Because we did not observe such correlation in sepsis, we may suggest a marginal role of this cytokine in monitoring the inflammatory status of these patients. Further, we can confirm that in patients with AOSD, IL-18 serum level correlates with disease activity, and its levels were significantly different between patients with active and inactive disease.

Such correlation of IL-18 with AOSD disease activity was reported by Kim, *et al*<sup>32</sup>, although in that study a better correlation was observed of another molecule, S100A8/A9, with serological and clinical variables. Currently, in the evaluation of AOSD disease activity, both Pouchot's and Rau's criteria have been used. We found that Rau's criteria were more sensitive in the detection of patients with active disease compared with Pouchot's criteria. This finding is further supported by the evidence of a significant difference between patients with active and inactive AOSD in ferritin serum level, a variable included in Rau's but not in Pouchot's criteria.

Our results confirm the key role of IL-18 in AOSD and we suggest the detection of this cytokine as a useful tool for differential diagnosis between AOSD and sepsis. Moreover, we suggest that Rau's criteria are more sensitive in defining AOSD disease activity.

## REFERENCES

1. Efthimiou P, Paik PK, Bielory L. Diagnosis and management of adult onset Still's disease. *Ann Rheum Dis* 2006;65:564-72.
2. Yamaguchi M, Ohta A, Tsunematsu T, Kasukawa R, Mizushima Y, Kashiwagi H, et al. Preliminary criteria for classification of adult Still's disease. *J Rheumatol* 1992;19:424-30.
3. Rosário C, Zandman-Goddard G, Meyron-Holtz EG, D'Cruz DP, Shoenfeld Y. The hyperferritinemic syndrome: macrophage activation syndrome, Still's disease, septic shock and catastrophic antiphospholipid syndrome. *BMC Med* 2013;11:185.
4. Bombardieri M, McInnes IB, Pitzalis C. Interleukin 18 as potential therapeutic target in chronic autoimmune/autoinflammatory conditions. *Expert Opin Biol Ther* 2007;7:31-40.
5. Colafrancesco S, Priori R, Alessandri C, Perricone C, Pendolino M, Picarelli G, et al. IL-18 serum level in Adult Onset Still's Disease:

- a marker of disease activity. *Intern J Inflammation* 2012;2012:156890.
6. Tschoeke SK, Oberholzer A, Moldawer LL. Interleukin-18: a novel prognostic cytokine in bacteria-induced sepsis. *Crit Care Med* 2006;34:1225-33.
  7. American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference: definitions for sepsis and organ failure, guide lines for the use of innovative therapies in sepsis. *Crit Care Med* 1992;20:864-74.
  8. Pouchot J, Sampalis JS, Beaudet F, Carette S, Décarry F, Salusinsky-Sternbach M, et al. Adult Still's disease: manifestations, disease course, and outcome in 62 patients. *Medicine* 1991;70:118-36.
  9. 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. 2010;37:2369-76.
  10. Priori R, Colafrancesco S, Gattamelata A, Di Franco M, Di Tondo U, Valesini G. Adult-onset still disease: a rare disorder with a potentially fatal outcome. *Autoimmunity Highlights* 2010;1:53-9.
  11. Pierrakos C, Vincent JL. Sepsis biomarkers: a review. *Crit Care* 2010;14:R15.
  12. Harbarth S, Holeckova K, Froidevaux C, Pittet D, Ricou B, Grau GE, et al. Diagnostic value of procalcitonin, interleukin 6 and interleukin 8 in critically ill patients admitted with suspected sepsis. *Am J Respir Crit Care Med* 2001;164:396-402.
  13. Scirè CA, Cavagna L, Perotti C, Bruschi E, Caporali R, Montecucco C. Diagnostic value of procalcitonin measurement in febrile patients with systemic autoimmune diseases. *Clin Exp Rheumatol* 2006;24:123-8.
  14. Kawashima M, Yamamura M, Taniai M, Yamauchi H, Tanimoto T, Kurimoto M, et al. Levels of interleukin-18 and its binding inhibitors in the blood circulation of patients with adult-onset Still's disease. *Arthritis Rheum* 2001;44:550-60.
  15. Kawaguchi Y, Terajima H, Harigai M, Hara M, Kamatani M. Interleukin-18 as a novel diagnostic marker and indicator of disease severity in adult-onset Still's disease. *Arthritis Rheum* 2001;44:1716-7.
  16. Sidiropoulos PI, Goulielmos G, Voloudakis GK, Petraki E, Boumpas DT. Inflammasomes and rheumatic diseases: evolving concepts. *Ann Rheum Dis* 2008;67:1382-9.
  17. Okamura H, Nagata K, Komatsu T, Tanimoto T, Nukata Y, Tanabe F, et al. A novel costimulatory factor for gamma interferon induction found in the livers of mice causes endotoxic shock. *Infect Immun* 1995;63:3966-72.
  18. Reddy P. Interleukin-18: recent advances. *Curr Opin Hematol* 2004;11:405-10.
  19. Conigliaro P, Priori R, Bombardieri M, Alessandri C, Barone F, Pitzalis C, et al. Lymph node IL-18 expression in adult-onset Still's disease. *Ann Rheum Dis* 2009;68:442-3.
  20. Priori R, Barone F, Alessandri C, Colafrancesco S, McInnes IB, Pitzalis C, et al. Markedly increased IL-18 liver expression in adult-onset Still's disease-related hepatitis. *Rheumatology* 2011;50:776-80.
  21. Grobmyer SR, Lin E, Lowry SF, Rivadeneira DE, Potter S, Barie PS, et al. Elevation of IL-18 in human sepsis. *J Clin Immunol* 2000;20:212-5.
  22. Endo S, Inada K, Yamada Y, Wakabayashi G, Ishikura H, Tanaka T, et al. Interleukin 18 (IL-18) levels in patients with sepsis. *J Med* 2000;31:15-20.
  23. Zaki Mel-S, Elgendy MY, El-Mashad NB, Farahat ME. IL-18 level correlates with development of sepsis in surgical patients. *Immunol Invest* 2007;36:403-11.
  24. Biet F, Loch C, Kremer L. Immunoregulatory functions of interleukin 18 and its role in defense against bacterial pathogens. *J Mol Med* 2002;80:147-62.
  25. Marshall JD, Aste-Amézaga M, Chehimi SS, Murphy M, Olsen H, Trinchieri G. Regulation of human IL-18 mRNA expression. *Clin Immunol* 1999;90:15-21.
  26. Oberholzer A, Steckholzer U, Kurimoto M, Trentz O, Ertel W. Interleukin-18 plasma levels are increased in patients with sepsis compared to severely injured patients. *Shock* 2001;16:411-4.
  27. Bozza FA, Salluh JL, Japiassu AM, Soares M, Assis EF, Gomes RN, et al. Cytokine profiles as markers of disease severity in sepsis: a multiplex analysis. *Crit Care* 2007;11:R49.
  28. Dinarello CA, Novick D, Kim S, Kaplanski G. Interleukin-18 and IL-18 binding protein. *Front Immunol* 2013;4:289.
  29. Emmanuilidis K, Weighardt H, Matevossian E, Heidecke CD, Ulm K, Bartels H, et al. Differential regulation of systemic IL-18 and IL-12 release during postoperative sepsis: high serum IL-18 as an early predictive indicator of lethal outcome. *Shock* 2002;18:301-5.
  30. Novick D, Schwartzburd B, Pinkus R, Suissa D, Belzer I, Sthoeger Z, et al. A novel IL-18BP ELISA shows elevated serum IL-18BP in sepsis and extensive decrease of free IL-18. *Cytokine* 2001; 14:334-42.
  31. Leng SX, McElhaney JE, Waltson JD, Xie D, Fedarko NS, Kuchel GA. ELISA and multiplex technologies for cytokine measurement in inflammation and aging research. *J Gerontol A Biol Sci Med Sci* 2008;63:879-84.
  32. Kim HA, An JM, Nam JY, Jeon JY, Suh CH. Serum S100A8/A9, but not follistatin-like protein 1 and interleukin 18, may be a useful biomarker of disease activity in adult-onset Still's disease. *J Rheumatol* 2012;39:1399-406.