Clinical Manifestations but not Cytokine Profiles Differentiate Adult-onset Still's Disease and Sepsis

MONIKA RAU, MARTIN SCHILLER, STEFAN KRIENKE, PETRA HEYDER, HANNES LORENZ, and NORBERT BLANK

ABSTRACT. ***Objective.*** To analyze clinical manifestations, serum ferritin, and serum cytokine levels in patients with adult-onset Still's disease (AOSD) or bacterial sepsis and to evaluate their potential use for differential diagnosis.

Methods. Twenty-two consecutive patients with the first flare of AOSD and 6 patients with an established diagnosis of AOSD under immunosuppressive therapy were compared with 14 patients with bacterial sepsis. Clinical manifestations were scored in a Pouchot AOSD activity score including elevated serum ferritin levels to obtain a modified Pouchot score. Serum cytokine profiles were analyzed from each patient.

Results. The scores of clinical manifestations using a modified Pouchot activity score were significantly higher in patients with active untreated AOSD (mean 5.60 ± 1.93) compared with patients with chronic AOSD (mean 1.16 ± 0.98; $p < 0.001$) and patients with sepsis (mean 2.38 ± 1.19; $p < 0.001$). A modified Pouchot score ≥ 4 shows a sensitivity of 92% and a specificity of 93% for active AOSD. Serum cytokine levels of interleukin 1ß (IL-1ß), IL-6, IL-8, IL-10, IL-12, IL-18, interferon-γ, tumor necrosis factor-α, and calprotectin were elevated in acute AOSD and sepsis. Significant differences were detected only in patients with sepsis who had higher levels of IL-6 and IL-8. The overlap of the 2 groups limits the use of cytokines for differential diagnosis in individual patients.

Conclusion. A modified Pouchot AOSD activity score including elevated serum ferritin levels was more useful to confirm the diagnosis of AOSD compared to patients with sepsis. Elevated serum cytokines correlate with inflammation but are of limited use to differentiate between active AOSD and bacterial sepsis. (J Rheumatol First Release September 1 2010; doi:10.3899/jrheum.100247)

Key Indexing Terms:
ADULT-ONSET STILL'S DISEASE | SEPSIS | INTERLEUKIN 1ß
INTERLEUKIN 6 | INTERLEUKIN 8 | CALPROTECTIN

An immediate diagnosis of patients presenting with systemic inflammatory disorders is still a great challenge in clinical practice. Differentiation between microbial infections and sepsis and rheumatic diseases such as adult-onset Still's disease (AOSD) remains especially difficult in many cases. AOSD is a rare, systemic inflammatory disorder of unknown origin occurring with spiking fever, evanescent rash, arthralgia or arthritis, sore throat, lymphadenopathy, hepatosplenomegaly, and myalgia[1,2]. In the acute phase, all organ systems can be affected and can present as pleuritis, pericarditis, acute respiratory distress syndrome, acute renal failure, acute liver failure, thrombotic thrombocytopenic purpura, or aseptic meningoencephalitis[2,3]. Laboratory tests show a strong inflammatory reaction associated with leukocytosis, anemia, increased levels of acute-phase reactants in serum, hyperferritinemia, and elevated hepatic enzymes[2,3,4]. Patients with AOSD can present with single or recurrent inflammatory attacks or with a persistent inflammatory reaction[5]. Recent studies have investigated cytokine signatures in patients with AOSD. Elevated interleukin 18 (IL-18) levels were found in acute and chronic AOSD and some studies proposed IL-18 as a diagnostic marker and indicator of disease severity[6,7,8,9]. IL-18 is produced by activated monocytes and is considered to orchestrate a type 1 T helper cell (Th1) response in patients with AOSD[6]. Further, activated monocytes secrete IL-6, IL-8, and tumor necrosis factor-α (TNF-α)[10,11] and Th1 cells secrete interferon-γ (IFN-γ), IL-2, and TNF-α[6,8], confirming the role of monocytes and Th1 cells in the pathogenesis of AOSD.

Patients with systemic microbial infections and sepsis present with distinct patterns in the ways that organs are affected[12]. Laboratory tests show nonspecific inflammation, and therapeutic decisions are based on clinical and laboratory findings with limited accuracy[13]. Recent studies investigated cytokine profiles including those of IL-1ß, IL-6, IL-8, IL-10, TNF-α, and procalcitonin to identify patients with

From the Division of Rheumatology, Internal Medicine 5, University of Heidelberg, Heidelberg, Germany.

M. Rau, MD; M. Schiller, MD; S. Krienke, Technical Assistant; P. Heyder, Technical Assistant; H. Lorenz, MD, Professor; N. Blank, MD, Assistant Professor, Division of Rheumatology, Internal Medicine 5, University of Heidelberg.

Address correspondence to Dr. N. Blank, University of Heidelberg – Rheumatology, Im Neuenheimer Feld 410, Internal Medicine 5, Heidelberg 69120, Germany.
E-mail: norbert.blank@med.uni-heidelberg.de

Accepted for publication June 8, 2010.

microbial sepsis. Elevated levels of IL-1ß, IL-6, TNF-α, and IL-10 have been described in several studies[14,15]. Serum levels of IL-8 and IL-18 were also elevated in patients with sepsis[14,16,17]. Therefore, neither inflammatory measurements nor organ dysfunction, nor the use of single biomarkers alone, can sufficiently differentiate between microbial sepsis and an autoinflammatory condition imitating sepsis. Some studies suggested procalcitonin as a biomarker for infections with gram-negative bacteria[18,19], but procalcitonin was also elevated in patients with AOSD[20], Wegener's granulomatosis[21], Behcet's disease[22], and Kawasaki disease[23] in the absence of an infection.

Calprotectin is produced by macrophages and neutrophils and is also known as a heterodimer of the myeloid-related proteins (MRP)-8 and MRP-14[24,25]. Calprotectin belongs to the S100 protein family and is also referred to as S100 A8/A9. Increased serum calprotectin levels have been found in rheumatoid arthritis (RA)[26], Crohn's disease[27], and multiple sclerosis[28]. In patients with juvenile idiopathic arthritis, calprotectin has been suggested as a marker for disease activity[26]. Recent *in vitro* and *in vivo* studies have shown that calprotectin might be involved in dysregulation of cardiac contractility in systemic inflammatory disease[29], indicating that calprotectin could be an indicator or mediator of multiorgan failure associated with sepsis.

We investigated cytokine profiles in patients with AOSD and sepsis to identify crucial mediators in the pathomechanism and to identify biomarkers that could be used for the differential diagnosis.

MATERIALS AND METHODS

Patients and healthy controls. Twenty-eight patients with AOSD were treated at the university hospital of Heidelberg between 2006 and 2008. All patients fulfilled the AOSD classification criteria by Yamaguchi, *et al*[30]. Serum samples from 18 patients with AOSD were available for cytokine analysis. The results were separately analyzed in patients with acute untreated disease (aAOSD; n = 12) and with chronic disease activity (cAOSD; n = 6) according to clinical measurements and elevated serum C-reactive protein (CRP) levels. AOSD activity was assessed by a modified Pouchot score as published, with 1 modification[3]. Recently, serum ferritin was proposed as a marker of disease activity[31] and the Fautrel classification criteria of AOSD included glycosylated ferritin[32]. Therefore, we included serum ferritin to obtain a modified Pouchot score[3] comprising 12 disease manifestations: fever, evanescent rashes, sore throat, arthritis, myalgia, pleuritis, pericarditis, pneumonitis, lymphadenopathy, hepatomegaly or abnormal liver function tests, elevated leukocyte count > 15,000/μl, and serum ferritin > 3000 μg/l.

The sepsis group consisted of 14 patients with systemic microbial infection who were treated at the intensive care units at the university hospital in Heidelberg. All patients in the sepsis group met the sepsis definition criteria of the American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference in 1991[33]. Blood samples were collected during the first 3 days of hospitalization.

Seven healthy blood donors were included in this study as a control group. All patients gave written informed consent. Acquisition and storage of serum samples was approved by the local ethics committee at the University of Heidelberg.

Cytokine assessment. Serum samples were collected between 9 AM and 11 AM to minimize diurnal variations of serum cytokines. Samples were processed within 1 hour and serum aliquots were stored at –70°C. FlowCytomix Multiplex (Bender MedSystems, Vienna, Austria) was used for detection of serum levels of IL-1ß, IL-2, IL-4, IL-5, IL-6, IL-8, IL-10, IL-12, IL-18, IFN-α, IFN-γ, TNF-α, and TNF-ß, according to the manufacturer's instructions. A commercial ELISA test was used for the measurement of calprotectin (PromoCell, Heidelberg, Germany).

Statistical methods. The Wilcoxon signed-rank test was employed for comparison of serum cytokine levels. The correlation coefficient was obtained by nonparametric Spearman's rank correlation test. P values < 0.05 were considered statistically significant.

RESULTS

Clinical characteristics. Clinical data were available from 15 male and 13 female patients with AOSD with a mean age (± SD) at disease onset of 32.2 ± 16.0 years. Patients with AOSD presented with daily high spiking fever (n = 26), arthritis/arthralgia (n = 27), evanescent rash (n = 16), elevated leukocyte count (n = 22), pharyngitis (n = 9), splenomegaly (n = 6), and elevated liver enzymes (n = 14) at disease onset. All 28 patients with AOSD received pulses of systemic glucocorticosteroids. In addition, some patients were treated with methotrexate (n = 20), leflunomide (n = 4), azathioprine (n = 2), cyclophosphamide (n = 1), and sulfasalazine (n = 1). Patients with chronic AOSD were treated with the biologics anakinra (n = 7), infliximab (n = 4), adalimumab (n = 3), and etanercept (n = 2) during the course of the disease. The clinical manifestations of all 28 patients with AOSD at the time of blood draw are shown in Table 1. The modified Pouchot activity score[3] was significantly higher in patients with aAOSD (mean = 5.60 ± 1.93) compared with patients with cAOSD (mean = 1.16 ± 0.98; p < 0.001) and patients with sepsis (mean = 2.38 ± 1.19; p < 0.001; Table 1), confirming that the modified Pouchot activity score is a useful clinical tool for differentiation between acute and chronic courses of AOSD and sepsis.

Serum samples were available from 18 of 28 patients with AOSD in our AOSD cohort. Twelve previously untreated patients with a flare and with elevated inflammatory measurements (CRP > 20 mg/l) were considered as aAOSD. Six patients with long-lasting disease received therapy with 1 or more immunosuppressive drugs. These patients had normal inflammatory measurements (CRP < 5 mg/l) and were considered as having cAOSD.

Fourteen patients with bacterial sepsis were chosen as a control group because of similar clinical symptoms. Six female and 8 male patients with a mean age (± SD) of 60.9 ± 16.8 years and serum CRP levels > 75 mg/l were included in our study. Eight patients with sepsis had pneumonia or were immunocompromised because of hematologic malignancies (n = 4). Three patients acquired sepsis after autologous blood stem cell transplant and were included after engraftment of blood stem cells. Six of 14 patients died during hospitalization. Serum samples from all 14 patients with sepsis and from 7 healthy blood donors (5 men, 2 women, mean age 55.1 ± 8.4 yrs) were acquired and cytokine profiles were analyzed.

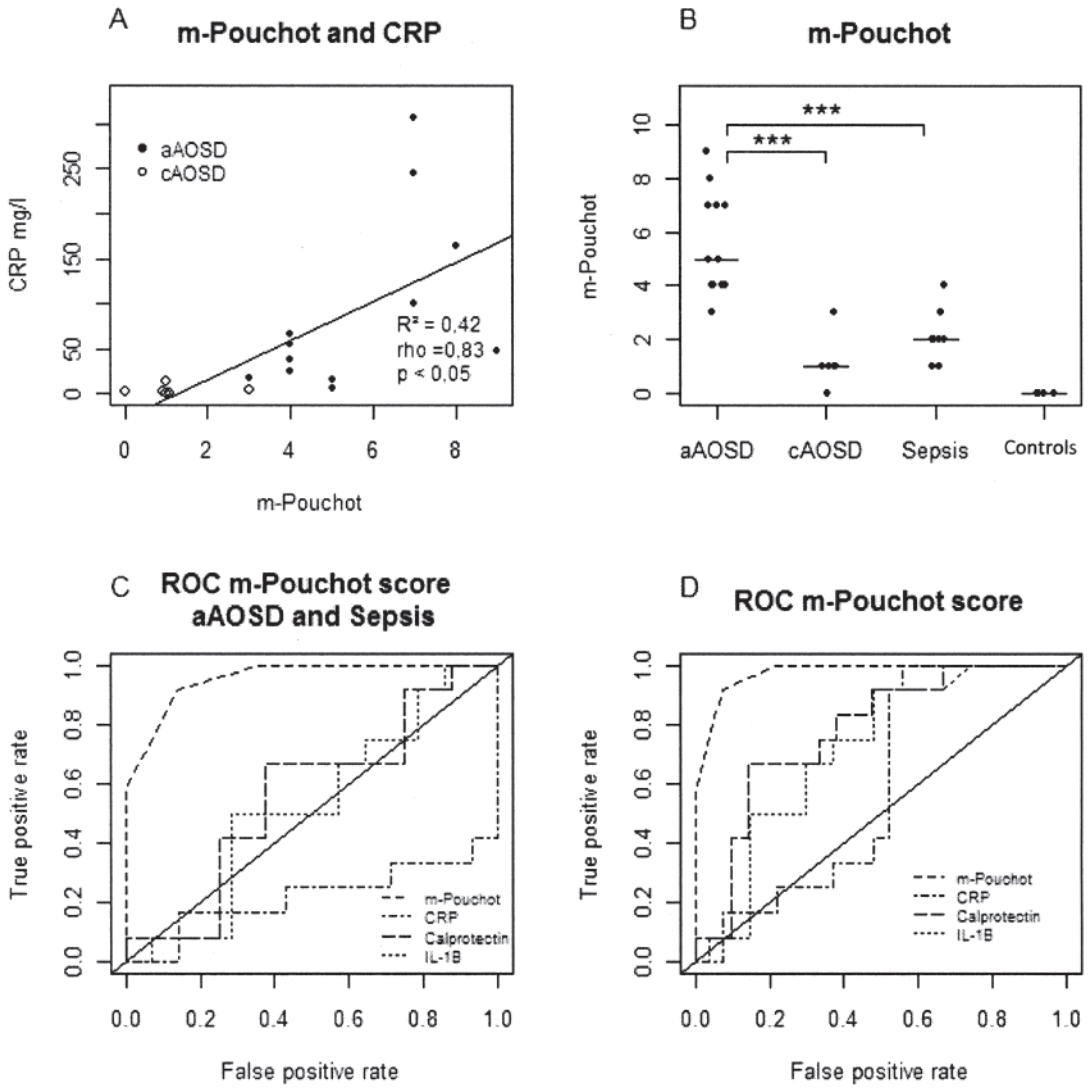

Figure 1. The modified Pouchot adult-onset Still's disease (AOSD) activity score correlates with C-reactive protein (CRP) levels in patients with acute or chronic AOSD (A) and differentiates active disease (aAOSD), inactive chronic disease (cAOSD), and patients with sepsis (B). Receiver-operation characteristic (ROC) curves for the modified Pouchot score and serum markers in patients with active AOSD and bacterial sepsis (C) and in all patients and blood donors (D) show a good discrimination of the modified Pouchot score but limited sensitivity and specificity of serum markers for the differential diagnosis between AOSD and sepsis. ***$p < 0.001$.

Routine laboratory test results. Twelve patients with aAOSD had anemia with Hb (mean ± SD) of 10.8 ± 2.1 g/dl, elevated leukocyte count 20,700 ± 5,100/μl, normal thrombocyte count, and elevated CRP levels of 102.8 ± 98.4 mg/l. Seven of 12 patients with aAOSD had elevated serum ferritin levels > 3000 μg/l, mean 10,327.7 ± 17,039.3 μg/l, at the time of the blood draw. Liver enzymes were elevated [glutamate pyruvate transaminase (GPT) 260.7 ± 433.4 U/l, glutamate oxalacetate transaminase (GOT) 632.4 ± 1136.8 U/l, lactate dehydrogenase (LDH) 563.2 ± 515.8 U/l] in 8 patients with aAOSD and 2 patients with cAOSD. CRP levels were 4.4 ± 4.8 mg/l in 6 patients with cAOSD.

An additional 10 patients with aAOSD but without available serum samples were included in this cohort and were analyzed from a clinical perspective and routine laboratory findings. During the acute phase of their disease, these patients with aAOSD had anemia with Hb 11.4 ± 1.8 g/dl, leukocyte counts of 14,330 ± 5,600/μl, normal thrombocyte counts, CRP levels of 123.6 ± 104.1 mg/l, and serum ferritin levels of 4,606.0 ± 12,396.7 μg/l. Liver enzymes were elevated in 6 of 10 patients with aAOSD (GPT 150.0 ± 330.9 U/l, GOT 223.7 ± 698.7 U/l, LDH 392.1 ± 402.9 U/l).

All 14 patients with sepsis had elevated CRP levels (157.2 ± 127.0 mg/l), anemia (Hb 9.6 ± 1.4 g/dl), and thrombocytopenia (103,400 ± 69,300/μl). Eight of 14 patients with sepsis had elevated leukocyte counts > 13,000/μl, and

Table 1. Clinical conditions of patients with adult-onset Still's disease (AOSD).

Clinical Conditions	Acute AOSD, n = 22	Chronic AOSD n = 6	Sepsis, n = 14
Fever	20	2	12
Evanescent rash	10	0	0
Pharyngitis	4	2	0
Arthritis	20	3	0
Myalgia	10	3	0
Pleuritis	7	0	2
Pericarditis	5	0	0
Pneumonitis	4	0	8
Lymphadenopathy	3	1	1
Hepatomegaly or elevated liver enzymes	11	0	2
Leukocyte count > 15,000/μl	14	0	5
Ferritin > 3000 μg/l	19	1	1
Mean (SD)	5.56 (1.67)	1.17 (0.98)	2.36 (0.93)
Median	5.00	1.00	2.00
p		< 0.001	
p			< 0.001

5 patients with sepsis were leukopenic with < 4000/μl. Although leukopenia was not associated with low cytokine levels, patients with sepsis < 1500 leukocytes/μl were excluded from our study. Healthy donors had normal laboratory results.

Modified Pouchot scores. Modified Pouchot scores and CRP levels showed a significant correlation in patients with aAOSD and cAOSD ($R^2 = 0.42$, rho = 0.83, $p < 0.05$; Figure 1A). The variation of the modified Pouchot score in individual patients shows a minimal overlap of both groups (Figure 1B). Therefore, a modified Pouchot score ≥ 4 can be used as a discrimination threshold for patients with active AOSD and bacterial sepsis. Receiver-operation characteristic (ROC) curves indicate that the modified Pouchot score has a high sensitivity and specificity for the diagnosis of active AOSD and for the differential diagnosis between active AOSD or sepsis (Figure 1C). In addition, ROC curves of the modified Pouchot score also indicate a high sensitivity and specificity for active AOSD compared with chronic-inactive AOSD and sepsis (Figure 1D). A modified Pouchot score cutoff value ≥ 4 predicted the correct diagnosis with good sensitivity (92%), specificity (93%), a negative predictive value of 96%, a positive predictive value of 85%, and an area under the ROC curve of 0.98 (Table 2). Other serum markers including IL-6, IL-8, IL-10, IL-12p70, IL-18, TNF-α, and IFN-γ show even lower sensitivities and specificities (data not shown) and are not useful for differentiation between active AOSD and sepsis.

Serum cytokine profiles in patients with AOSD and sepsis. Serum samples were available from 18 patients with AOSD (12 aAOSD, 6 cAOSD), 14 patients with sepsis, and 7 healthy blood donors. IL-1ß (Figure 2A) and IL-8 (Figure 2C) were detected in almost all sera, but IL-6 (Figure 2B) and IL-10 (Figure 2D) were barely detectable in healthy donors. IL-1ß, IL-6, and IL-10 levels were up to 10 pg/ml and IL-8 levels up to 230 pg/ml in healthy donors and patients with cAOSD (Figure 2).

Serum cytokine levels for IL-1ß, IL-6, IL-8, and IL-10 were elevated in patients with aAOSD and sepsis but not in patients with cAOSD or healthy donors (Figure 2 A–D). Concentrations of IL-1ß were detected up to ~1000 pg/ml, and IL-6, IL-8, and IL-10 were detected up to 11,647 pg/ml. The highest levels of IL-6 and IL-8 were detected in patients with sepsis (Figure 2). Also, medians of IL-6 and IL-8 were significantly higher in patients with sepsis compared to patients with aAOSD (Figure 2B, 2C; $p < 0.01$).

Table 2. Sensitivity, specificity, predictive values, and area under the receiver-operation characteristic curve (AUC ROC) for modified Pouchot score and serologic measurements.

Factor	Cutoff Value	Sensitivity, %	Specificity, %	NPV, %	PPV, %	AUC ROC
Modified Pouchot score	≥ 4	92	93	96	85	0.98
CRP	15.6 mg/l	92	48	93	44	0.59
IL-1ß	10.9 pg/ml	92	52	93	46	0.72
IL-6	7.2 pg/ml	83	48	87	42	0.56
IL-8	40.6 pg/ml	75	41	79	36	0.44
Calprotectin	3.6 ng/ml	83	62	87	56	0.77

NPV: negative predictive value; PPV: positive predictive value; CRP: C-reactive protein; IL: interleukin.

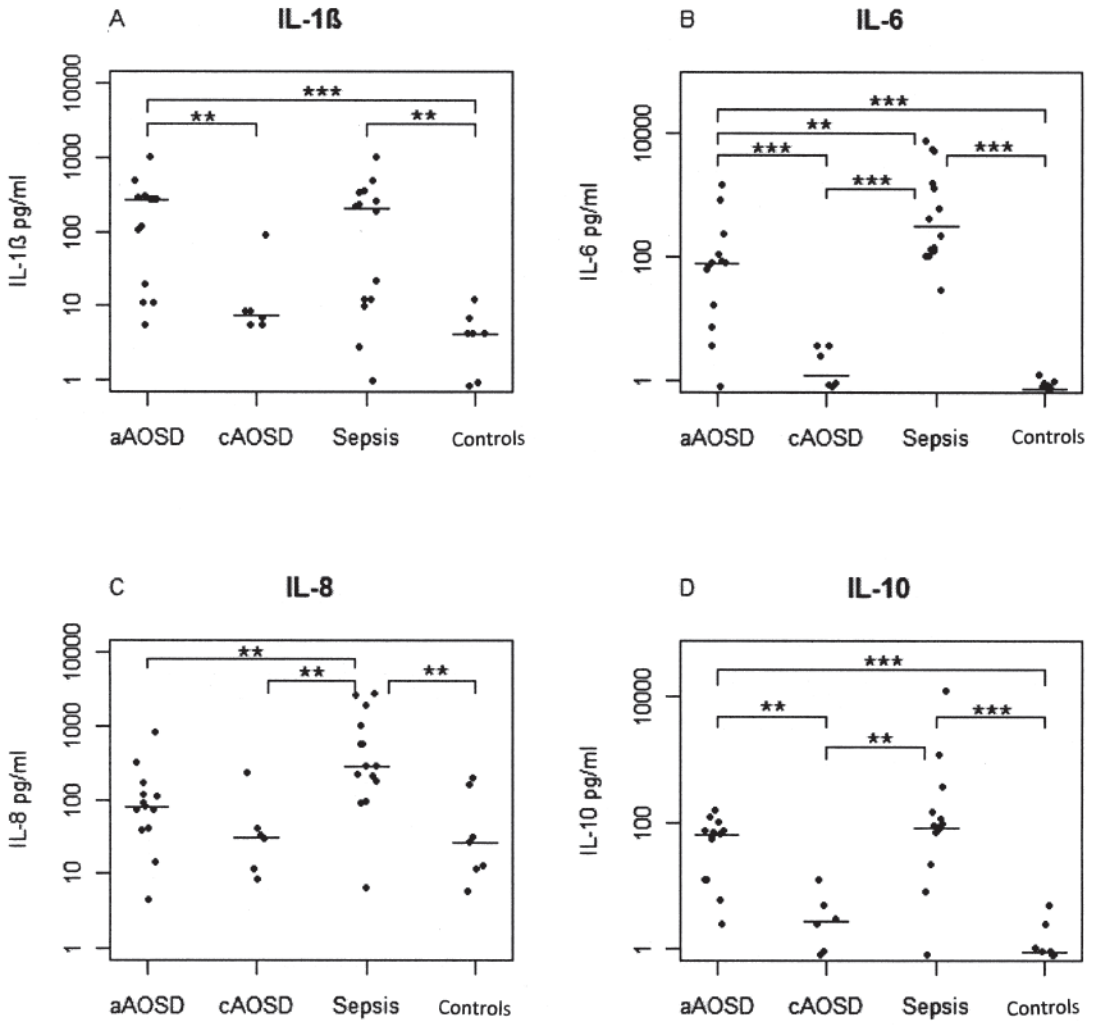

Figure 2. Serum levels of interleukin (IL)-1ß, IL-6 (B), IL-8 (C), and IL-10 (D) in patients with active adult-onset Still's disease (aAOSD; n = 12), chronic disease activity (cAOSD; n = 6), sepsis (n = 14), and in controls (n = 7). Medians are shown as horizontal lines. Statistical analyses were performed using a Mann-Whitney U test. **p < 0.01, ***p < 0.001.

Our data indicated that patients with sepsis could be differentiated from patients with aAOSD by higher levels of IL-6 and IL-8. However, although differences were statistically significant, we observed a broad overlap of cytokine levels in single patients (Figures 2B, 2C), compromising the usefulness of these cytokines in individual patients.

The monocyte-derived cytokines IL-12p70 (Figure 3A), TNF-α (Figure 3C), and calprotectin (Figure 3D), and the Th1 lymphocyte-derived IFN-γ (Figure 3B) showed a similar distribution with high serum levels in patients with aAOSD and sepsis and moderate or low levels in patients with cAOSD and healthy donors. Calprotectin was detected in sera at concentrations between 500 and 9000 pg/ml (Figure 3D). Significantly elevated calprotectin levels were detected in patients with aAOSD and sepsis with no significant difference between the 2 groups (Figure 3D).

We did not detect any significant differences of these cytokines between patients with aAOSD and patients with sepsis.

The highest concentrations of the monokine IL-18 were detected up to ~200,000 pg/ml in individual patients with aAOSD but also in patients with sepsis (data not shown). However, because of the broad range of IL-18 levels in all groups of patients, we did not find significant differences between patients with active AOSD and those with sepsis (data not shown).

DISCUSSION

Patients with AOSD exhibit a combination of several disease manifestations. An accurate diagnostic procedure requires consideration and exclusion of differential diagnoses and recognition of typical clinical and laboratory findings of AOSD. The clinical and laboratory features of bacterial sepsis can be similar to those of active AOSD. Our

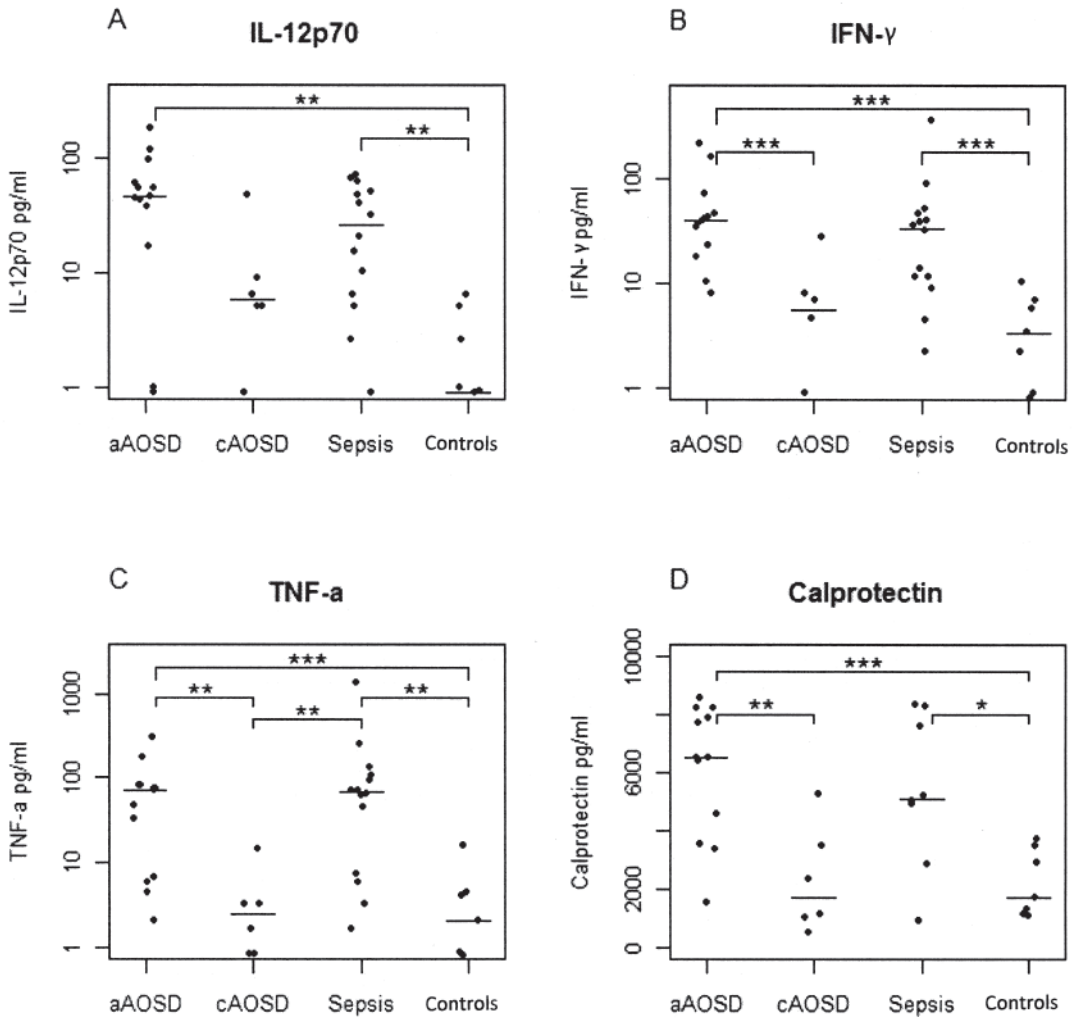

Figure 3. Serum levels of IL-12p70 (A), interferon-γ (IFN-γ) (B), tumor necrosis factor-α (TNF-α) (C), and calprotectin (D) in patients with aAOSD, cAOSD, sepsis, and controls. Medians are shown as horizontal lines. Statistical analyses were performed using the Mann-Whitney U test. *p < 0.05, **p < 0.01, ***p < 0.001.

results show that a modified Pouchot score combines information about clinical symptoms and laboratory results and shows a minimal overlap in individual patients. Therefore, a modified Pouchot score can differentiate between AOSD and bacterial sepsis. A modified Pouchot cutoff score ≥ 4 shows a sensitivity of 92% and a specificity of 93% for the diagnosis of AOSD.

We investigated whether serum cytokine levels can contribute to the differential diagnosis between AOSD and bacterial sepsis. A plethora of studies investigated cytokine profiles in patients with AOSD[6,7,8,9,10,11] and bacterial infection[12,13,14,15,16,17,18,19], but to our knowledge this is the first study that directly compares serum cytokines in patients with active AOSD or bacterial sepsis. Elevated monocyte-derived cytokines such as IL-6, IL-8, IL-18, and TNF-α[6,7,8,9,10,11] and Th1-derived cytokines such as IFN-γ indicated that activated monocytes and activated Th1 cells probably have a crucial role in the pathogenesis of AOSD. However, similar results were also reported in patients with bacterial sepsis[12,14,15,16,17,18,19]. These results were confirmed by our data. Significantly higher levels of IL-6 and IL-8 were detected in sera from patients with sepsis compared to active AOSD. However, because of the broad overlap of the 2 groups, these cytokines are of limited use for the differential diagnosis in clinical practice. Other cytokines did not show significant differences between patients with AOSD and patients with sepsis.

Activated monocytes secrete IL-1ß, IL-6, IL-8, IL-18, and TNF-α. Synthesis of IL-1 and IL-18 requires activation of Toll-like receptors and nuclear factor-κB as well as activation of intracellular Nod-like receptors and subsequent interaction of an adaptor-mediated multiprotein complex called the inflammasome. The inflammasome activates caspase-1, which cleaves inactive proleukines into biologically

active IL-1 and IL-18, which could be secreted by monocytes[34]. IL-1ß is the principal pyrogenic cytokine[34] that might be responsible for the daily spiking fever in patients with aAOSD. Intrinsic overstimulation of IL-1ß production is associated with autoinflammatory syndromes, which show similar clinical signs during a flare compared with patients with AOSD or sepsis. Significantly elevated levels of IL-1ß were also detected in patients with active disease, which is consistent with previous reports in patients with aAOSD[34] or sepsis[14,35]. Because large numbers of IL-1ß receptors are abundant in all tissues, unbound IL-1ß has a very short lifetime *in vivo*[34], indicating that we probably detect only a small amount of total IL-1ß. An intrinsic overproduction of IL-1ß can be reduced with drugs such as anakinra, rilonacept, or canakinumab. However, these biologic drugs probably enhance immunosuppression in patients with sepsis, which might cause death. Therefore, a rapid and reliable differentiation between AOSD and sepsis is mandatory before the initiation of anti-IL-1ß targeted therapy.

Calprotectin is another biomarker that is produced by activated monocytes. Good correlations between calprotectin and CRP and other acute-phase reactants were reported recently in patients with juvenile idiopathic arthritis[26], polymyalgia rheumatica[36], and RA[37]. Our data show that patients with aAOSD have significantly higher serum calprotectin levels than patients with cAOSD, but calprotectin levels cannot differentiate between microbial and intrinsic inflammation.

Previous studies suggested that hypoglycosylated serum ferritin (below 20% of total serum ferritin) could help to differentiate AOSD from other inflammatory conditions[38]. However, the same report showed that hypoglycosylated serum ferritin can also be detected in patients who are non-AOSD but have bacterial infections or systemic disease[38], indicating overlapping conditions. In addition, glycosylated serum ferritin tests were not routinely available in many institutions. Another recent report suggested the use of procalcitonin to differentiate between autoinflammatory disease activity and bacterial infection in patients with AOSD[39]. However, our experience with procalcitonin indicated a limited specificity of procalcitonin for bacterial infections in patients with autoimmune or autoinflammatory diseases. Therefore, procalcitonin was not determined routinely in our institution.

Some limitations of our study are the low number of patients and the restriction to 13 cytokines. Cytokines such as IL-1ß, IL-6, and TNF-α are known to exhibit diurnal variations. In addition, detection of cytokines can be compromised by soluble cytokine receptors and a short half-life *in vivo*[40,41]. To minimize these effects, we standardized our sample preparation procedure as indicated, but we cannot fully exclude these variations from our study.

Our data show that the modified Pouchot score comprising typical clinical signs and laboratory findings can differentiate between patients with AOSD and patients with bacterial sepsis. Serum cytokines are elevated in both conditions and are of limited use to differentiate between AOSD and bacterial sepsis.

REFERENCES

1. Bywaters EG. Still's disease in the adult. Ann Rheum Dis 1971;30:121-33.
2. Kontzias A, Efthimiou P. Adult-onset Still's disease: pathogenesis, clinical manifestations and therapeutic advances. Drugs 2008;68:319-37.
3. Pouchot J, Sampalis JS, Beaudet F, Carette S, Decary F, Salusinsky-Sternbach M, et al. Adult Still's disease: manifestations, disease course, and outcome in 62 patients. Medicine 1991;70:118-36.
4. Pouchot J, Vinceneux P. [Clinical and biological manifestations of adult-onset Still's disease]. Presse Med 2004;33:1012-8.
5. Efthimiou P, Paik PK, Bielory L. Diagnosis and management of adult onset Still's disease. Ann Rheum Dis 2006;65:564-72.
6. Chen DY, Lan JL, Lin FJ, Hsieh TY, Wen MC. Predominance of Th1 cytokine in peripheral blood and pathological tissues of patients with active untreated adult onset Still's disease. Ann Rheum Dis 2004;63:1300-6.
7. Choi JH, Suh CH, Lee YM, Suh YJ, Lee SK, Kim SS, et al. Serum cytokine profiles in patients with adult onset Still's disease. J Rheumatol 2003;30:2422-7.
8. Fujii T, Nojima T, Yasuoka H, Satoh S, Nakamura K, Kuwana M, et al. Cytokine and immunogenetic profiles in Japanese patients with adult Still's disease. Association with chronic articular disease. Rheumatology 2001;40:1398-404.
9. Kawaguchi Y, Terajima H, Harigai M, Hara M, Kamatani N. Interleukin-18 as a novel diagnostic marker and indicator of disease severity in adult-onset Still's disease. Arthritis Rheum 2001;44:1716-7.
10. Chen DY, Lan JL, Lin FJ, Hsieh TY. Proinflammatory cytokine profiles in sera and pathological tissues of patients with active untreated adult onset Still's disease. J Rheumatol 2004;31:2189-98.
11. Hoshino T, Ohta A, Yang D, Kawamoto M, Kikuchi M, Inoue Y, et al. Elevated serum interleukin 6, interferon-gamma, and tumor necrosis factor-alpha levels in patients with adult Still's disease. J Rheumatol 1998;25:396-8.
12. Schuetz P, Christ-Crain M, Muller B. Biomarkers to improve diagnostic and prognostic accuracy in systemic infections. Curr Opin Crit Care 2007;13:578-85.
13. Vincent JL, Abraham E. The last 100 years of sepsis. Am J Respir Crit Care Med 2006;173:256-63.
14. Bozza FA, Salluh JI, 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.
15. Marchant A, Deviere J, Byl B, De Groote D, Vincent JL, Goldman M. Interleukin-10 production during septicaemia. Lancet 1994;343:707-8.
16. Chollet-Martin S, Montravers P, Gibert C, Elbim C, Desmonts JM, Fagon JY, et al. High levels of interleukin-8 in the blood and alveolar spaces of patients with pneumonia and adult respiratory distress syndrome. Infect Immun 1993;61:4553-9.
17. 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.
18. 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.

19. Muller B, Schuetz P, Trampuz A. Circulating biomarkers as surrogates for bloodstream infections. Int J Antimicrob Agents 2007;1:S16-23.
20. Scire 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.
21. Moosig F, Csernok E, Reinhold-Keller E, Schmitt W, Gross WL. Elevated procalcitonin levels in active Wegener's granulomatosis. J Rheumatol 1998;25:1531-3.
22. Adam B, Calikoglu E. Serum interleukin-6, procalcitonin and C-reactive protein levels in subjects with active Behcet's disease. J Eur Acad Dermatol Venereol 2004;18:318-20.
23. Okada Y, Minakami H, Tomomasa T, Kato M, Inoue Y, Kozawa K, et al. Serum procalcitonin concentration in patients with Kawasaki disease. J Infect 2004;48:199-205.
24. Striz I, Trebichavsky I. Calprotectin — a pleiotropic molecule in acute and chronic inflammation. Physiol Res 2004;53:245-53.
25. Yui S, Nakatani Y, Mikami M. Calprotectin (S100A8/S100A9), an inflammatory protein complex from neutrophils with a broad apoptosis-inducing activity. Biol Pharm Bull 2003;26:753-60.
26. Frosch M, Strey A, Vogl T, Wulffraat NM, Kuis W, Sunderkotter C, et al. Myeloid-related proteins 8 and 14 are specifically secreted during interaction of phagocytes and activated endothelium and are useful markers for monitoring disease activity in pauciarticular-onset juvenile rheumatoid arthritis. Arthritis Rheum 2000;43:628-37.
27. Aadland E, Fagerhol MK. Faecal calprotectin: a marker of inflammation throughout the intestinal tract. Eur J Gastroenterol Hepatol 2002;14:823-5.
28. Bogumil T, Rieckmann P, Kubuschok B, Felgenhauer K, Bruck W. Serum levels of macrophage-derived protein MRP-8/14 are elevated in active multiple sclerosis. Neurosci Lett 1998;247:195-7.
29. Feuerstein GZ. Cardiac RAGE in sepsis: call TOLL free for anti-RAGE. Circ Res 2008;102:1153-4.
30. 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.
31. Van Reeth C, Le Moel G, Lasne Y, Revenant MC, Agneray J, Kahn MF, et al. Serum ferritin and isoferritins are tools for diagnosis of active adult Still's disease. J Rheumatol 1994;21:890-5.
32. Fautrel B, Zing E, Golmard JL, Le Moel G, Bissery A, Rioux C, et al. Proposal for a new set of classification criteria for adult-onset still disease. Medicine 2002;81:194-200.
33. American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference: definitions for sepsis and organ failure and guidelines for the use of innovative therapies in sepsis. Crit Care Med 1992;20:864-74.
34. Dinarello CA. The IL-1 family and inflammatory diseases. Clin Exp Rheumatol 2002;20:S1-13.
35. Calandra T, Baumgartner JD, Grau GE, Wu MM, Lambert PH, Schellekens J, et al. Prognostic values of tumor necrosis factor/cachectin, interleukin-1, interferon-alpha, and interferon-gamma in the serum of patients with septic shock. Swiss-Dutch J5 Immunoglobulin Study Group. J Infect Dis 1990;161:982-7.
36. Brun JG, Madland TM, Gran JT, Myklebust G. A longitudinal study of calprotectin in patients with polymyalgia rheumatica or temporal arteritis: relation to disease activity. Scand J Rheumatol 2005;34:125-8.
37. Hammer HB, Odegard S, Fagerhol MK, Landewe R, van der Heijde D, Uhlig T, et al. Calprotectin (a major leucocyte protein) is strongly and independently correlated with joint inflammation and damage in rheumatoid arthritis. Ann Rheum Dis 2007;66:1093-7.
38. Fautrel B, Le Moël G, Saint-Marcoux B, Taupin P, Vignes S, Rozenberg S, et al. Diagnostic value of ferritin and glycosylated ferritin in adult onset Still's disease. J Rheumatol 2001;28:322-9.
39. Chen DY, Chen YM, Ho WL, Chen HH, Shen GH, Lan JL. Diagnostic value of procalcitonin for differentiation of bacterial infection and non-infectious inflammation in febrile patients with active adult-onset Still's disease. Ann Rheum Dis 2009;68:1075.
40. Damas P, Ledoux D, Nys M, Vrindts Y, De Groote D, Franchimont P, et al. Cytokine serum level during severe sepsis in human IL-6 as a marker of severity. Ann Surg 1992;215:356-62.
41. Hildebrand F, Pape HC, Krettek C. [The importance of cytokines in the posttraumatic inflammatory reaction]. Unfallchirurg 2005;108:793-4, 796-803.