



Utility of neutrophil CD64 and serum TREM-1 in distinguishing bacterial infection from disease flare in SLE and ANCA-associated vasculitis

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Abstract

Bacterial and opportunistic infections are a major cause of morbidity and mortality in patients with systemic lupus erythematosus (SLE) and antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis owing to treatment with immunosuppressants. Commonly used laboratory tests are unreliable in differentiating infection from active disease patients. Fc receptor (FcγR1 or CD64) expression on neutrophils and soluble TREM-1 (triggering receptor expressed on monocytes) are potential biomarkers of bacterial infections. Our aim was to measure the clinical usefulness of quantitative CD64 measurement on neutrophils and soluble TREM-1 measurements in differentiating bacterial infection from active disease in patients with SLE and ANCA vasculitis. Patients with bacterial infection ($n = 25$), active disease ($n = 51$), and healthy controls ($n = 20$) were included. Neutrophil CD64 expression using flow cytometry and sTREM-1 and procalcitonin levels by ELISA were studied. The percentage of neutrophils with CD64 expression and their mean fluorescence intensity in patients with infection (68.8 (56.9–86.5)%, 1037 (229–1828)) were significantly ($p < 0.05$) higher as compared to those without infection (7.7 (2.6–13.1)%, 456 (20–968)) and controls (7.05 (1.4–9.5)%, 99.5 (54.7–140.7)). The sensitivity and specificity of CD64 expression on neutrophils to diagnose bacterial infection (using a cutoff value of 30%) was 85% and 84%, respectively, whereas the sensitivity and specificity of procalcitonin was 75% and 85%, respectively. There was no significant difference in soluble TREM-1 levels between the two groups. Quantitative measurement of CD64 on neutrophils can distinguish between systemic infection and the flare of autoimmune diseases.

Keywords AAV · ANCA-associated vasculitis · Biomarker · CD64 · Sepsis · SLE

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Abbreviations

AAV	ANCA-associated vasculitis
ANCA	Anti-neutrophil-associated cytoplasmic antibody
ELISA	Enzyme-linked immunosorbent assay
ESR	Erythrocyte sedimentation rate
MAS	Macrophage activation syndrome
MFI	Mean fluorescence intensity
PBS	Phosphate-buffered saline
PMN	Polymorphonuclear leukocytes
RA	Rheumatoid arthritis
SLE	Systemic lupus erythematosus
TREM	Triggering receptor expressed on monocytes

Introduction

Patients with rheumatic diseases are predisposed to infection because of the disturbances caused in the immune system by the disease itself as well as by the various immunosuppressive drugs the patients are on. Such infections are a major cause of mortality in systemic lupus erythematosus (SLE) and antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis (AAV) [1, 2]. Differentiation between a flare of a rheumatic disease and systemic infection in a patient receiving immunosuppressive treatment is vitally important as the treatment differs dramatically [3]. However, in routine clinical practice, this remains a common dilemma since many patients present with signs and symptoms of non-specific inflammation like fever, arthralgia, and shortness of breath that can be caused by both disease activity and infection.

The complexity of the clinical presentation of both infections and active disease creates the need of biomarker that can reliably differentiate between the two. While such biomarkers are well described to distinguish active from inactive disease [4], there is a paucity of biomarkers to distinguish active disease from infection. Common screening laboratory tests employed to diagnose infections, such as total leukocyte number, presence of immature forms in peripheral smear, C-reactive protein (CRP), and erythrocyte sedimentation rate (ESR), have poor sensitivity and specificity [5]. Culture results are often viewed as confirmatory, but in practice, they are often not used in immediate treatment decisions because of their relatively slow turnaround times of up to 72 h or more. Research in both soluble biomarker and quantitative cellular measurements promises the availability of improved diagnostic techniques. One such candidate is neutrophil CD64 (FcγRI). Multiple studies have proven the utility of neutrophil CD64 (nCD64) as a marker of sepsis, but it remains unclear whether CD64 can be useful to distinguish infection from active disease in a patient with SLE and AAV, since these diseases are per se associated with activation of the immune system. Few studies have reported positive results with higher levels of CD64 in patients with infection as compared to active

disease [6, 7]. However, the patient numbers were limited [5]. Another potential biomarker is sTREM-1 (soluble triggering receptor expressed on myeloid cells type 1), which has been evaluated as a marker of sepsis [8, 9]. It has been studied in SLE [10, 11] and AAV [12] to identify patients with infection. In the present study, we have evaluated the clinical usefulness of quantitative CD64 measurements on neutrophils and sTREM-1 in serum to differentiate between systemic infection and active autoimmune inflammation in patients with SLE and AAV.

Material and methods

This cross-sectional observational descriptive study was conducted in the Department of Clinical Immunology, Sanjay Gandhi Post-graduate Institute of Medical Sciences, Lucknow, India. The study was approved by the institutional ethics committee.

Patients

All diagnosed patients of SLE (fulfilling SLICC (Systemic Lupus International Collaborating Clinics) criteria [13]) and AAV (using European Medicines Agency algorithm [14]) admitted to clinical immunology ward with suspected active disease or bacterial infection were prospectively recruited in the study after obtaining written informed consent. A total of 76 patients and 20 healthy controls were included. Neutropenic patients (absolute neutrophil count less than 1500/μl), patients having received G-CSF (granulocyte-colony stimulating factor) therapy, and patients suspected to have mycobacterial or fungal infection by the treating physician were excluded. Six milliliters of venous blood (3 ml plain + 3 ml in ethylenediaminetetraacetic acid (EDTA)) was collected from each patient and from healthy controls.

Patients were divided into three groups—active disease, proven infection, and probable infection. Active disease was defined as patients with SLEDAI (SLE disease activity index) >4 and BVAS (Birmingham Vasculitis Activity Score) >0 with no feature to suggest infection. Patients in the infection group were those in whom the pathogen was proven by microbiological culture. Patients with probable infection were those who had obvious symptoms of infection such as pus discharge, strong suspicion based on radiological findings such as lung abscess on CT chest or those in whom infection was assumed due to response to antibiotics. For the purpose of comparison, patients with proven and probable infection were analyzed together and compared with those with active disease.

Neutrophil CD64 expression

CD64 expression on neutrophils was analyzed from whole blood. One hundred microliters of EDTA blood was incubated with 20 μ l PE (phycoerythrin)-conjugated human anti-CD64 antibody (BD Pharmingen, CA, USA) and 20 μ l PE-conjugated matched-isotype control antibody for 30 min at room temperature. Then, RBCs were lysed with BD FACS lysing solution and washed twice in phosphate-buffered saline (PBS.) Afterwards, 500 μ l of PBS was added finally to the tube and acquired on BD FACS Canto flow cytometer (BD Biosciences). At least 10,000 cells were counted and analyzed and separated according to their forward and side scatter characteristics. Cell population showing mean fluorescence intensity (MFI) that indicated positive staining for corresponding CD64 marker was identified by comparison of dual-parameter histograms to the corresponding negative control. The positive region or an individual sample was based on the non-immune mouse IgG PE, with the percentage of cells in the positive region of this negative control sample generally less than 2% of total events. Results are expressed as the percentage of neutrophils expressing CD64 [15] and as mean fluorescence intensity (MFI) [16].

sTREM-1 and procalcitonin

Blood samples were centrifuged at 1500 rpm for 10 min, serum aliquoted, and stored at -80°C until analysis. sTREM-1 and procalcitonin levels were measured in duplicate after a single freeze-thaw cycle in batched assays by sandwich enzyme-linked immunosorbent assay (ELISA) as per manufacturer's protocol. ELISA kit from R&D systems (catalog no. DY1278B, DY8350-05), respectively, were used. Analytical sensitivity of the immunoassay for s-TREM1 was 93.75–6000 pg/ml and procalcitonin was 31.3–2000 pg/ml.

Lab parameters

CRP and complements (C3, C4) in the serum were measured by nephelometry. Complement levels were measured only in patients of SLE. anti-MPO (myeloperoxidase), and anti-PR3 (proteinase-3) levels were measured by ELISA while ESR was measured by Westergren method.

Statistical analysis

All data was analyzed using SPSS 20 software. CD64 expression and sTREM-1 levels were compared by non-parametric tests (Mann-Whitney *U* test) between active disease and infection group. Intergroup comparisons were carried out using non-parametric tests. Multivariate analysis was performed using logistic regression analysis. Receiver operator characteristic (ROC) curve analysis was used to establish optimal

sensitivity and specificity of the CD64 assay. A *p* value of < 0.05 was considered significant.

Results

Patients

A total of 76 patients were recruited. Demographic data is provided in Table 1. The autoimmune features present in SLE active and infection groups are provided in supplementary Table 1. Among them, 51 were admitted with active disease and 25 were admitted with infection. Among the 51 patients with active disease, 35 had SLE and 16 had AAV. Among the 25 patients with infection, 21 had underlying SLE while 4 had AAV. Seventy-three percent of patients in active disease group and 72% of infection group had fever at presentation. Among the infection group, 14 had culture proven infection, while 11 had probable infection. Site of infections were lung, skin, urinary tract, and dental. In the patients with proven infection, the organisms, which were isolated, were *E. coli*, *Staphylococcus*, *Klebsiella*, *Pneumococcus*, and *Pseudomonas*.

Neutrophil CD64 expression

Percentage of neutrophils expressing CD64 and MFI was significantly higher in patients with infection as compared to active disease and healthy controls on multivariate logistic regression analysis (Table 2, Fig. 1) ($p < 0.05$). Neutrophil CD64 was the only variable observed to be significant in multivariate analysis. CD64 expression was similar in healthy controls and those with active disease. With ROC curve analysis nCD64% of 30 was detected to have a sensitivity of 85% and a specificity of 84% in differentiating disease activity with bacterial infection with an area under the curve of 0.87 (on combining both SLE and AAV patients' data). The positive and negative predictive values were 72.4% and 91.5%, respectively. There was no correlation between duration of fever and nCD64%.

Two SLE patients with macrophage activation syndrome had markedly elevated nCD64%. These two patients were not included in the final analysis.

sTREM-1

sTREM-1 was higher in patients with disease activity and infection in comparison to health controls (Table 2). However, sTREM-1 levels were not significantly different between patients with disease activity and infection.

Table 1 Demographic and clinical characteristics of patients with SLE and AAV with and without infection

Characteristic	SLE with active disease (n = 35)	SLE with infection (n = 21)	AAV with active disease (n = 16)	AAV with infection (n = 4)	Healthy controls (n = 20)
Age in years	26 (20–31)	28 (24–41)	41 (35–62)	35 (27–47)	25 (22–26)
Gender—female (%)	94%	100%	56%	50%	60%
Duration of underlying illness in months	24 (6–63)	10 (3.5–19.5)	4 (2–6)	30 (12–54)	
SELENA-SLEDAI	14 (8–16)	7.5 (0.5–16)	NA	NA	
Patients with SLEDAI \geq 4	35 (100%)	18 (85%)	NA	NA	
BVAS	NA	NA	13 (9.5–17)	4.5 (0–9)	
Fever (%)	26 (74%)	16 (76%)	11 (69%)	2 (50%)	
Temperature on admission (°F)	100.4 (98.8–101.2)	101.4 (100–102.3)	101.2 (99.4–102)	99.6 (98.9–102.2)	
Serum creatinine	0.95 (0.8–1.4)	1.1 (0.8–1.2)	1.1 (0.75–1.7)	1.5 (1.35–1.8)	
Organ involvement					
Lung ^a	2	11	8	2	
Kidney	20	9	9	4	
ENT	Nil	Nil	7	1	
Gastrointestinal system	3	4	1	1	
Nervous system	3	1	5	Nil	

Values have been expressed as median (25th–75th interquartile range)

NA not applicable

^a Excluding pleural effusion

Procalcitonin

Procalcitonin was higher in patients with infection compared to those with disease activity. At a cutoff value of 244 pg/ml, it had a sensitivity and specificity of 75% and 85% respectively, to differentiate between the two, with an area under the curve of 0.75 (on combining both SLE and AAV patients' data). The positive and negative predictive values were 70.4% and 87.8%, respectively.

Other lab parameters

Total leukocyte count (TLC), ESR, CRP, C3, C4, anti-dsDNA, anti-MPO, and anti-PR3 were not significantly different in patients with active disease and infection.

Combining tests

If either a high nCD64% or procalcitonin was taken to indicate infection, then sensitivity increased to 97%; however, specificity fell to 71.4%. If increased levels of both were taken to

Table 2 Comparison of various parameters between healthy controls, patients with active disease and infection

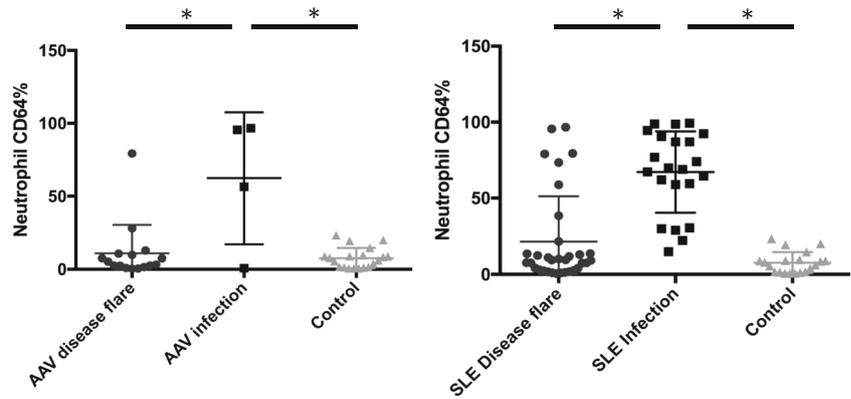
Characteristic	Active disease (n 49)	Infection (n 25)	Healthy controls (n 20)
CD64 expression on neutrophils (%)	7.7 (2.6–13)	68.8 (56.9–86.5)*	7.05 (1.4–9.5)
Mean fluorescence intensity of CD64	456 (20–986)	1037 (229–1828)*	99.5 (54.7–140.7)
sTREM-1 (pg/ml)	1184 (717–1609)**	899 (531–1284)**	255.1 (95.1–634.8)
Procalcitonin (pg/ml)	157.7 (115.1–209.3)**	301 (141.8–339.3)*	77.6 (61–101.8)
TLC (4000–11,000 cells/mm ³)	9700 (4600–12,400)	10,750 (6000–15,000)	–
CRP (< 5 mg/L)	1.9 (0.86–6.05)	5.4 (1.83–8.9)	–
ESR (< 20 mm/h)	70 (30–103)	75 (55–110)	–

All values have been expressed as median (25th–75th interquartile range)

CRP C-reactive protein, ESR erythrocyte sedimentation rate, TLC total leukocyte count, sTREM soluble triggering receptor expressed on myeloid cells

* $p < 0.05$ vs. active disease group and healthy controls; ** $p < 0.05$ vs. healthy controls

Fig. 1 CD64 expression (number of neutrophils expressing CD64). **a** ANCA vasculitis patients and controls. **b** SLE patients and controls



indicate infection, sensitivity and specificity were 64% and 97.6%, respectively.

Subgroup analysis

On subgroup analysis of patients with underlying SLE, procalcitonin, TLC, and CRP were observed to be significantly higher in patients with SLE with bacterial infection compared to those with active disease (Table 3). However, for patients with SLE, CD64 had a higher sensitivity and specificity, and a higher area under the curve on ROC analysis in comparison with procalcitonin, CRP, and TLC (Fig. 2). If either a high nCD64% or procalcitonin is taken to indicate infection, then the sensitivity increases to 99%; however, the specificity falls to 70%. If increased levels of both are taken to indicate infection sensitivity falls to 76% and specificity increases to 97%. Among the 21 SLE patients with infections, the admitting physician admitted 7 patients suspecting disease activity. Out of these seven, five had raised nCD64 (> 30) at admission.

On subgroup analysis of patients with AAV, there was no difference in procalcitonin, CRP, and TLC in patients with and without infection.

Discussion

We measured neutrophil CD64 expression and sTREM-1 levels in 76 patients of SLE and AAV (compared with 20 healthy controls) who had presented with either disease activity or infection. We found higher nCD64 in patients with infection compared to active disease, with 85% sensitivity and 84% specificity in differentiating between the two at a cutoff value of 30. It had better area under the curve than procalcitonin. sTREM-1 levels and other lab parameters (CRP, ESR, TLC) were not different between active disease and infection group. On subgroup analysis of SLE patients, although CRP and TLC were significantly different in infection and active disease group, they had a lower sensitivity and specificity in comparison to nCD64 expression. The higher

Table 3 Subgroup analysis of patients with SLE and ANCA-associated vasculitis

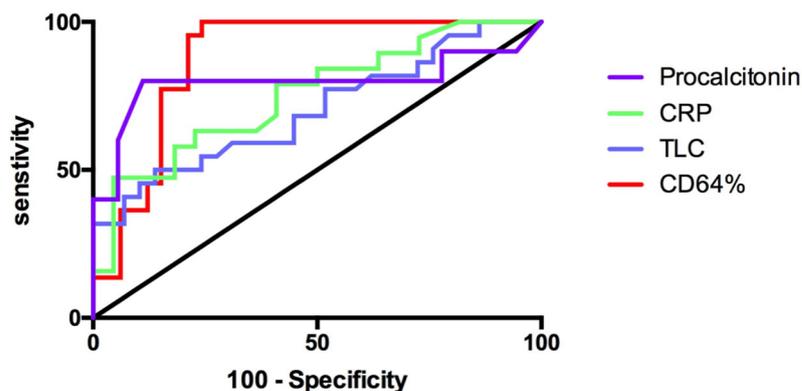
Variable	SLE with active disease (n = 33)	SLE with infection (n = 21)	AAV with active disease (n = 16)	AAV with infection (n = 4)
CRP (mg/dl)	1.3 (0.39–2.3)	5.54 (1.8–9.2)*	7.3 (2.64–10.5)	11.7 (2.08–20.85)
TLC (cells/mm ³)	6400 (3400–9500)	11,000 (5925–16,650)*	12,200 (10,550–20,400)	12,000 (10,300–20,100)
nCD64 (%)	9.42 (3–13.7)	69.4 (59–87.1)*	4.25 (2.27–8.5)	76 (42.5–80.9)#
Procalcitonin (< 150 pg/ml)	174 (101–204)	316 (231–345)*	172 (127–257)	104 (86–356)
sTREM-1 (pg/ml)	1231 (764–1663)	897 (430–1284)	1015 (244–1493)	1272 (810–2154)
Anti-dsDNA (IU/ml)	300 (107–427)	153 (87–380)	–	–
C3 (60–120 mg/dl)	60.2 (36.3–91.9)	52.2 (36.3–79.5)	–	–
C4 (15–25 mg/dl)	14.6 (6.6–19)	17.4 (10.4–36.9)	–	–
ESR (< 20 mm/h)	60 (27–94)	95 (70–113)	88 (66–123)	32.5 (21–85)

All values have been expressed as median (25th–75th interquartile range)

AAVANCA-associated vasculitis, CRP C-reactive protein, TLC total leukocyte count, nCD64 number of neutrophils expressing CD64, C3 complement 3, C4 complement 4

*p < 0.05 vs. SLE with disease flare; # p < 0.05 vs. AAV with disease activity

Fig. 2 ROC analysis of various parameters in patients with underlying SLE. nCD64 number of neutrophils expressing CD64, CRP C-reactive protein, TLC total leukocyte count, PPV positive predictive value, NPV negative predictive value



Marker	Area under curve	Cut-off	Sensitivity	Specificity	PPV	NPV
nCD64 (%)	0.88	22	95%	80%	74%	96.5%
Procalcitonin (pg/ml)	0.79	254	80%	88%	80.9%	88.6%
CRP (mg/dl)	0.75	1.6	76%	60%	53.3%	80.7%
TLC (cells/mm ³)	0.7	10000	50%	78%	55.6%	73%

level of CRP in patients with infection is predictable since CRP is usually normal in patients with SLE [17]. We also found that in five patients in whom the admitting physician suspected disease flare but later were found to have infection, nCD64% could have given a clue.

The Fc receptors, member of immunoglobulin supergene family, are expressed on white blood cells, where they function to integrate response involving both the innate and acquired immune systems. CD64 is the Fc receptor that binds with monomeric IgG type antibodies with high affinity. It is more commonly known as Fc γ receptor I (Fc γ RI). It is constitutively expressed on macrophages, monocytes, and eosinophils. Normally, CD64 is present on the surface of a few circulating polymorphonuclear leukocytes (PMN); however, neutrophil CD64 expression rapidly increases as a physiological response to microbial wall components, complement split products, and some pro-inflammatory cytokines within 4 to 6 hours [18–20]. It has been reported to be a good biomarker for early diagnosis of bacterial infection [21]. With respect to SLE patients, increased CD64 expression has been observed on circulating monocytes, but these levels were not elevated on neutrophils [22]. In a study including 19 patients with SLE and 24 with rheumatoid arthritis (RA), the nCD64

was significantly higher than in the infection group compared to those with active disease, while there was no significant difference between the group of patients with inactive inflammatory disease and healthy controls. At a cutoff of 43.5%, nCD64 had a 94.4% sensitivity and 88.9% specificity to detect an infection in patients with SLE and RA [6]. In a similar study of 44 patients with various autoimmune disease (RA, 21; SLE, 8; gout, 8; vasculitis, 5; others, 7), the sensitivity and specificity of CD64 expression on neutrophils to diagnose systemic infection was 85% and 91%, respectively [23]. These results are similar to what we observed.

Utility of nCD64 has not been well studied in patients with ANCA vasculitis. Though neutrophils have been shown to be activated in AAV and express CD64, whether it can differentiate with infection, has not been studied well in previous studies. In a study of 28 patients with AAV, nCD64 was significantly higher in AAV patients compared to controls. However, there was no difference in nCD64 levels in AAV patients with quiescent and active disease. On comparison with patients with sepsis (without underlying AAV), the authors observed higher nCD64 expression in those with sepsis compared to active AAV [24]. Since fever, raised leukocyte count, and CRP cannot reliably differentiate between infection

and disease activity in AAV, nCD64 is a potential biomarker to differentiate between the two in AAV patients.

An interesting observation was the elevated levels of nCD64% in SLE patients with macrophage activation syndrome (MAS). This has not been reported before and requires further research. In patients with SLE with leucopenia, high-grade fever, elevated nCD64%, and sterile cultures should be suspected to have macrophage activation. Therefore, nCD64% could be unreliable to differentiate between SLE patients with MAS and those with infection. MAS could have been precipitated by an infection as well, and in the setting of MAS with lupus, to differentiate disease activity from infection, there is a need to seek further biomarkers.

TREM-1 has been identified on the surface of mature neutrophils and monocytes responding to bacterial and fungal infections. Its engagement leads to amplification of innate immune response by activation of NF- κ B. Its production is regulated by lipoteichoic acid and bacterial lipopolysaccharide [25]. Increase in membrane expression of TREM-1 is also associated with the release of the soluble form sTREM-1 in sepsis [8]. In a study of 98 patients with SLE and 49 healthy controls, sTREM-1 levels were observed to be elevated in SLE patients with infection as compared to healthy subjects (1.1 vs. 0.11 pg/ml). In the same study, SLE patients with signs of active disease without infection were reported to have a higher value of sTREM-1 compared with both healthy subjects and SLE patients without signs of activity (2.9 vs. 0.8 pg/ml, respectively) [10]. However, when patients with infection (n 20) were compared to those without infection, there was no difference in levels of sTREM-1 levels, similar to our results. Conflicting results have been reported in a study in which sTREM-1 levels were studied in 32 SLE (33 febrile episodes—19 infections and 14 flares) patients complaining of fever. sTREM-1 levels on the day of recruitment of patients were significantly higher in the infection group than in the flare group (109.9 pg/ml (median) vs. 48.0 pg/ml, $p = 0.002$) [11]. The conflicting result could be due to the difference in inclusion criteria, since only those patients who complained of fever or were recorded to have fever during a hospital admission were recruited.

In a previous study of 41 patients with anti-MPO renal vasculitis, sTREM-1 was measured in 29 samples from patients with active disease, 27 patients with inactive disease, and 17 samples from AAV patients with infection. There was significant correlation between serum levels of soluble TREM-1 and serum creatinine levels among all patients ($r = 0.554$, $p < 0.0001$). The serum-soluble TREM-1/creatinine ratio was higher in inactive vasculitis patients with infectious complications than in active vasculitis, inactive vasculitis without infection, and chronic kidney disease (CKD) patients ($p = 0.0005$, $p < 0.0001$, and $p < 0.0001$, respectively), but not significantly different to that in acute pyelonephritis patients [12]. The correlation between sTREM-1 and serum creatinine

was not seen in our study, and neither was there a difference in sTREM-1/creatinine ratio between the two groups (not shown). Again, the difference in results could be due to the difference in patient profile, since only six AAV patients in our study had renal failure while most patients in this study carried out by a nephrology unit had rapidly progressive renal failure. In another study of 12 patients of AAV, it was reported that sTREM-1 level was elevated during the active phase compared to the inactive phase in the same patients on follow-up (0.27 ng/ml (median) vs. 0.06 ng/ml (median), $p < 0.05$) [26]. sTREM-1 needs to be studied further in SLE and AAV patients before any definite conclusion can be drawn.

During the time period of our study, there was an epidemic of dengue. We measured nCD64 expression in 20 dengue patients within first 5 days of disease onset and found the median nCD64 expression to be 34% (24–39). This suggests that nCD64 expression is elevated even in patients with viral infection, although not to levels as high as seen in bacterial infections. We also repeated nCD64 expression in six patients with infection on the day of discharge (median hospital stay, 5 days) after they had been treated with antibiotics and found it to be decreased (37 (29–44)%) compared to the level on the day of admission (73 (68–86)%).

Procalcitonin, a proven marker of bacterial sepsis has also been evaluated in detecting bacterial infection in patients with underlying autoimmune disease. It was found to be more sensitive and specific than CRP in the detection of bacterial infections in 44 patients with autoimmune diseases, including 10 SLE patients [27]. In a meta-analysis, it had a higher diagnostic value than CRP in the detection of bacterial sepsis in patients with autoimmune diseases, with a sensitivity of 75% and a specificity of 90% [28]. In a study of 114 SLE patients (67 without and 47 with infection), it was reported that a value > 380 pg/ml had a 74.5% sensitivity and a 95.5% specificity, a high positive predictive value of 92.1% and a negative predictive value of 84.2% for the differentiation of infection and disease activity [29]. These results are similar to ours.

On subgroup analysis, we found that procalcitonin levels were not different between AAV patients with and without infection. This is in contrast to previous studies in which procalcitonin was found to be higher in patients with infection compared to those with disease activity in AAV [30]. In a study with 53 AAV patients (43 without and 10 with infection), procalcitonin was more useful than CRP with the best cutoff at 0.1 ng/ml (sensitivity 60%, specificity 92%) [27]. The conflicting results could be due to the fact that we had a lesser number of AAV patients with infection. It is important to reiterate that procalcitonin also rises in non-septic inflammatory conditions such as after surgical procedures or after resuscitation, in those with cardiogenic shock, or with severe systemic inflammatory states such as severe pancreatitis or rhabdomyolysis [31].

A limitation of our study is the small number of AAV patients recruited especially in the AAV with infection group. Also, we have not carried out serial testing of nCD64% in patients receiving antibiotics (those with infection) or immunosuppressives (those with active disease).

In conclusion, we propose that nCD64 expression may be a useful tool in those patients with SLE and AAV who present with non-specific symptoms such as fever, suggesting either flare of the underlying disease or systemic infection. It might be even more useful in AAV since unlike SLE, CRP and TLC are elevated in patients with AAV with active disease. It is a simple test with a short turnaround time (1–2 h) and has a comparable cost with CRP and procalcitonin (about 5 USD per assay in our laboratory). This test can be performed at any laboratory with flow cytometry facilities and does not need other special equipment or expertise. However, each laboratory would need to establish its own cutoff values. These characteristics make it an attractive test to incorporate into daily clinical practice. To the best of our knowledge, we report the highest number of SLE and AAV patients in which nCD64 has been studied to differentiate between infection and active disease. The role of sTREM-1 needs to be studied further to see if it can be used as a biomarker of infection in patients with SLE and AAV, although in our study, it was not found to be useful.

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Compliance with ethical standards

Disclosures None.

Ethical approval The pertinent ethical committee has given its approval and/or the reported investigations have been performed in accordance with the principles of the Declaration of Helsinki.

Informed consent Informed consent had been obtained from the patients, whenever appropriate.

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