OBSERVATIONAL STUDY

OPEN

Persistently Elevated Soluble Triggering Receptor Expressed on Myeloid Cells 1 and Decreased Monocyte Human Leucocyte Antigen DR Expression Are Associated With Nosocomial Infections in Septic Shock Patients

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OBJECTIVES: Sepsis-acquired immunosuppression may play a major role in patients' prognosis through increased risk of secondary infections. Triggering receptor expressed on myeloid cells 1 (TREM-1) is an innate immune receptor involved in cellular activation. Its soluble form (sTREM-1) has been described as a robust marker of mortality in sepsis. The objective of this study was to evaluate its association with the occurrence of nosocomial infections alone or in combination with human leucocyte antigen-DR on monocytes (mHLA-DR).

DESIGN: Observational study.

SETTING: University Hospital in France.

PATIENTS: One hundred sixteen adult septic shock patients as a post hoc study from the IMMUNOSEPSIS cohort (NCT04067674).

INTERVENTIONS: None.

MEASUREMENTS AND MAIN RESULTS: Plasma sTREM-1 and monocyte HLA-DR were measured at day 1 or 2 (D1/D2), D3/D4, and D6/D8 after admission. Associations with nosocomial infection were evaluated through multivariable analyses. At D6/D8, both markers were combined, and association with increased risk of nosocomial infection was evaluated in the subgroup of patients with most deregulated markers in a multivariable analysis with death as a competing risk. Significantly decreased mHLA-DR at D6/D8 and increased sTREM-1 concentrations were measured at all time points in nonsurvivors compared with survivors. Decreased mHLA-DR at D6/D8 was significantly associated with increased risk of secondary infections after adjustment for clinical parameters with a subdistribution hazard ratio of 3.61 (95% CI, 1.39–9.34; $p = 0.008$). At D6/D8, patients with persistently high sTREM-1 and decreased mHLA-DR presented with a significantly increased risk of infection (60%) compared with other patients (15.7%). This association remained significant in the multivariable model (subdistribution hazard ratio [95% CI], 4.65 [1.98–10.9]; $p < 0.001$).

CONCLUSIONS: In addition to its prognostic interest on mortality, sTREM-1, when combined with mHLA-DR, may help to better identify immunosuppressed patients at risk of nosocomial infections.

KEY WORDS: human leucocyte antigen-DR on monocytes; immunosuppression; mortality; nosocomial infection; septic shock; soluble triggering receptor expressed on myeloid cells 1

Despite an improved understanding of the pathophysiology of sepsis, this syndrome remains a major cause of morbidity and mortality worldwide (1). The recent analysis of the Global Burden of Disease Study estimates

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KEY POINTS

Question: While elevated sTREM-1 plasma levels are associated with increased risk of death after sepsis, no study investigated its association with occurrence of nosocomial infections in adult septic shock patients.

Findings: In this observational study including 116 septic shock patients, we described that patients with persistently high sTREM-1 and decreased HLA-DR expression on monocytes at the end of the first week after ICU admission presented with a significantly increased risk of infection independently of usual clinical confounding factors.

Meaning: In addition to its prognostic interest on mortality, sTREM-1, when combined with mHLA-DR, may help to better identify immunosuppressed patients at risk of nosocomial infections.

that 48.9 million cases and 11 million deaths are related to sepsis each year worldwide (2). Due to aging population and better treated comorbidities, sepsis incidence will continue to rise, and related mortality remains high in northern countries (26.5% for hospitalized sepsis and 41.6% for septic shock) (3, 4).

Based on 2016 international definition (Sepsis-3), sepsis is now defined as a life-threatening organ dysfunction caused by a dysregulated host response to infection. This includes septic shock when vasopressor therapy is required to maintain mean arterial pressure of 65 mm Hg or greater, and serum lactate level greater than 2 mmol/L despite adequate volume resuscitation (5).

The immunoinflammatory host response is central to the pathophysiology of sepsis in a complex and overtime evolving phenomenon. Indeed, initial hyperinflammation leading to organ failure and early deaths concomitantly induces compensatory mechanisms (i.e., negative feedback to inhibit deleterious inflammation), that may, in a significant number of patients, become dominant and persist overtime. As a consequence, these patients present with a profound state of acquired immunosuppression (6, 7). As both innate and adaptive immunities are altered, this contributes to dampen immune surveillance as illustrated by increased rate of nosocomial infections (due to opportunistic germs), viral reactivation, or incapacity to clear the initial infectious site (4, 8–10). With improvement of organ support therapies, early detection of infections, and timely initiation of antimicrobials, more patients now survive the first initial hyperinflammatory phase of sepsis and may later develop sepsis-acquired immunosuppression. This second immunosuppressive phase of the disease is, thus, supposed to be associated with more than 70% of total sepsis-related mortality (4).

Therefore, immunotherapy has emerged as a promising therapeutic approach for restoring immune defenses in selected patients presenting with the most altered immune parameters (11–13). As there is no clinical sign of immunosuppression, this tailored therapeutic approach must be based on a biomarker-guided strategy. Among biomarkers available routinely, consensus now exists for considering low monocyte HLA-DR expression (human leucocyte antigen-DR on monocytes [mHLA-DR]) as the reference surrogate marker to identify immunosuppressed patients (14, 15).

Triggering receptor expressed on myeloid cells 1 (TREM-1) is an innate immunity receptor expressed by myeloid cells (i.e., neutrophils, macrophages, and monocytes) belonging to the immunoglobulin superfamily (16–18). Its activation leads to cytokine synthesis (interleukin [IL]-8, monocyte chemoattractant protein [MCP] 1 and MCP-3, and tumor necrosis factor **α**), neutrophil degranulation, and oxidative burst, and it contributes to optimize antigen presentation process by increasing CD80/CD86 expressions on monocytes (19–21). TREM-1 works in synergy with NOD-like receptor and toll-like receptor to amplify their responses (22, 23). It is a membrane receptor inducing intracytosolic activity through receptor dimerization and involvement of DNAX Activating Protein of 12 kD protein adaptor, which carries immunoreceptor tyrosine-based activation motifs patterns (24). Once activated, a soluble form of TREM-1 (sTREM-1) is released due to surface proteolysis by metalloproteases (25). Although sTREM-1 is believed to possess negative retrocontrol functions at cell level (25), it is also a specific marker of the activation of the TREM-1 pathway (26).

Described as a marker of the proinflammatory phase, sTREM-1 was initially studied as a diagnostic marker of infection, in blood or bronchoalveolar lavage (4, 27, 28). Thereafter, sTREM-1 was repeatedly

reported as a good marker for predicting mortality in septic patients (29–32). To our knowledge, no study has investigated the prognostic value of sTREM-1 regarding the risk of occurrence of nosocomial infections in adult septic shock patients.

The main objective of this study was, thus, to investigate the association between plasma sTREM-1 concentration alone or in combination with mHLA-DR and the occurrence of nosocomial infections in a cohort of septic shock patients.

MATERIAL AND METHODS

Design

We performed a post hoc retrospective analysis of sTREM-1 plasma level in a selected cohort of patients included in the IMMUNOSEPSIS study (NCT04067674), a prospective, single-center, observational cohort study conducted in the Anesthesiology and Intensive Care Department at the Edouard Herriot Hospital, Hospices Civils de Lyon, France. Clinical data (age, sex, length of stay, type of infection, biological evidence of immune response and impact of organ failure, exposure to invasive devices, living or deceased status after 28 d, and presence or absence of nosocomial infection after 28 d) were collected blindly from results of biological analyses by clinicians. Nosocomial infection diagnostic was made according to the definition of European Center for Disease Prevention and Control (available at: https://www.ecdc.europa.eu/en/all-topics/eu-case-definitions). All nosocomial infections were evaluated retrospectively, based on medical record, by two different physicians. Physicians remained blinded for results of immune parameters. Only the first episode of secondary infection was considered in the analyses. Agreement between physicians was obtained for all patients. mHLA-DR was measured at D1/D2 after admission in ICU (D1/D2), then between days 3 and 4 (D3/D4), and, finally, between days 6 and 8 (D6/D8). sTREM-1 measurements were performed at the same time points.

Patients

The inclusion criteria were patients admitted to ICU with septic shock and available plasma samples at D1/D2 and D3/D4. The exclusion criteria were any condition modifying the immune status: immunosuppressive treatment (including >10 mg equivalent prednisone per day or cumulative dose > 700 mg), hematological disease treated within the 5 years, solid tumor treated with chemotherapy or in remission, number of circulating neutrophils less than 500/mm^3, innate immune deficiency, and extracorporeal circulation with 1 month before inclusion (cardiopulmonary bypass or extracorporeal membrane oxygenation). Patients were selected by their chronological inclusion in IMMUNOSEPSIS.

Ethic Approval and Consent to Participate

Because of the noninterventional nature of the study, written informed consent was not required for inclusion in IMMUNOSEPSIS study. However, each clinician orally informed the patient or family members about the objectives, methodology, and conduct of the study, and provided them with a leaflet using plain language. Non opposition to participation to this study was systematically obtained from the patient or a third party before any blood sampling was performed and was recorded in patient's clinical file. All procedures performed were in accordance with the ethical standards of the institutional and/or national research committee (Comité de Protection des Personnes Sud-Est II, IRB11236) and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards. The biological samples were stored in the Sepsis Biological Collection located at the Immunology Laboratory, Edouard Herriot Hospital, Hospices Civils de Lyon. The biological collection was recorded at the Ministry of Health in August 2008, under number DC-2008-509. Clinical and biological databases were reported to the Commission nationale de l'informatique et des libertés—National Commission for Information Technology and Freedom (number 08-27).

Biomarker Measurements

mHLA-DR expression was measured on fresh whole blood using standardized flow cytometry technique. Cell staining was performed using Quantibrite reagents (antibodies and beads, Becton Dickinson, Franklin Lakes, NJ) as previously described (33, 34). Results were expressed as the number of antibodies bound per cell (AB/C).

sTREM-1 plasma level was determined by Enzyme-Linked Immunosorbent Assay (Human TREM-1 Quantikine ELISA Kit, R&D Systems, Minneapolis, MN).

Statistical Analysis

The data were described as numbers and percentages for categorical variables and medians and interquartile ranges for continuous variables. Comparisons between groups relied on the Fisher exact test or chi-square test for categorical data and on the Wilcoxon test for continuous data. The Kaplan-Meier analysis was performed, and log-rank test was applied. Associations between sTREM-1 or mHLA-DR measurements and D28-mortality were first analyzed. Then, we carried out the same approach regarding the occurrence of nosocomial infections. In this study, the occurrence of nosocomial infections was followed up until day 28 for all patients. Death was, thus, a competing event because it precludes the occurrence of nosocomial infection. Therefore, a competing risk analysis using a Fine and Gray model was used in order to specifically evaluate the association between immune parameters and occurrence of nosocomial infections (35). A regression on cumulative incidence functions was then realized. This multivariable analysis was conducted including variables significantly associated with infection occurrence in univariate analysis and clinically relevant variables. Age, Sequential Organ Failure Assessment (SOFA) score at admission, sepsis source, and invasive mechanical ventilation were tested in the model. The final multivariable model was selected using a stepwise method retaining only significant variables. Adjusted subdistribution hazard ratio (sHR) and their 95% CIs were calculated. Finally, in order to assess the potential of the combination of sTREM-1 and mHLA-DR at D6/D8 in regard with association with nosocomial infection occurrence, we defined four subgroups according to high or low biomarker values as defined as above or below medians. In that respect, patients with high sTREM-1 (above median) and low mHLA-DR (below median) were classified into an extreme phenotype (EP) group and were compared with all other patients. In this analysis, only patients alive after this screening were considered. All statistical tests were two sided, with $p < 0.05$ considered significant. Statistical analysis was computed with R software (Version 4.2.1; R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Patients

One hundred sixteen patients were included in the study. The median age was 71 years (62.5–79 yr). The median SOFA score at inclusion was 9 (7.25–11.0), and the median Simplified Acute Physiology Score II score was 58 (48.75–75.0). All patients needed vasoactive drugs, 81% required mechanical ventilation, and 17.2% renal replacement therapy. Primary site of infection was abdominal in 46% of patients, pulmonary in 19%, and urinary tract in 11.2%. Patients' baseline characteristics are reported in **Supplemental Table 1** (http://links.lww.com/CCX/B146). Of the 116 patients, a total of 29 (25 %) died during the first 28 days with a median delay of 9.5 days. Thirty-one (26.7%) patients presented with at least one nosocomial infection within 28 days after inclusion with a median delay of 10 days. Of these, 17 patients (54.8%) had ventilator-associated pneumonia, and 8 (25.8%) had a central venous catheter infection.

Association With Mortality

First, we aimed to confirm the association between elevated sTREM-1 plasma concentration and 28-day mortality after sepsis as previously described in other cohorts. As expected, during overall monitoring period (i.e., at D1/D2, D3/D4, and D6/D8), sTREM-1 levels were significantly higher at each time point in deceased septic patients compared with survivors (**Fig. 1*A***). As previously reported, lower mHLA-DR value expressions were associated with mortality only solely at the latest time point (i.e., D6/D8; **Fig. 1*B***).

Association With Nosocomial Infections

The clinical characteristics of patients who developed or not nosocomial infections are presented in **Table 1**. Regarding occurrence of nosocomial infections, only mHLA-DR expression at D6/D8 was significantly different between groups. Patients developing nosocomial infections presented with lower mHLA-DR values than noninfected patients (5662 AB/C and 7676 AB/C, respectively; $p = 0.03$) (**Fig. 2**). Cumulative incidence of nosocomial infections was significantly different between groups stratified on median mHLA-DR expression (i.e., 6668 AB/C; $p = 0.0065$), with death as a competing risk (**Fig. 3*A***). In the subgroup of patients with mHLA-DR at D6/8 below median, 44.4% developed at least one nosocomial infection compared with 15.3% in the group of patients with higher mHLA-DR. In multivariable analysis, after adjustment based on age and SOFA score, low mHLA-DR expression at

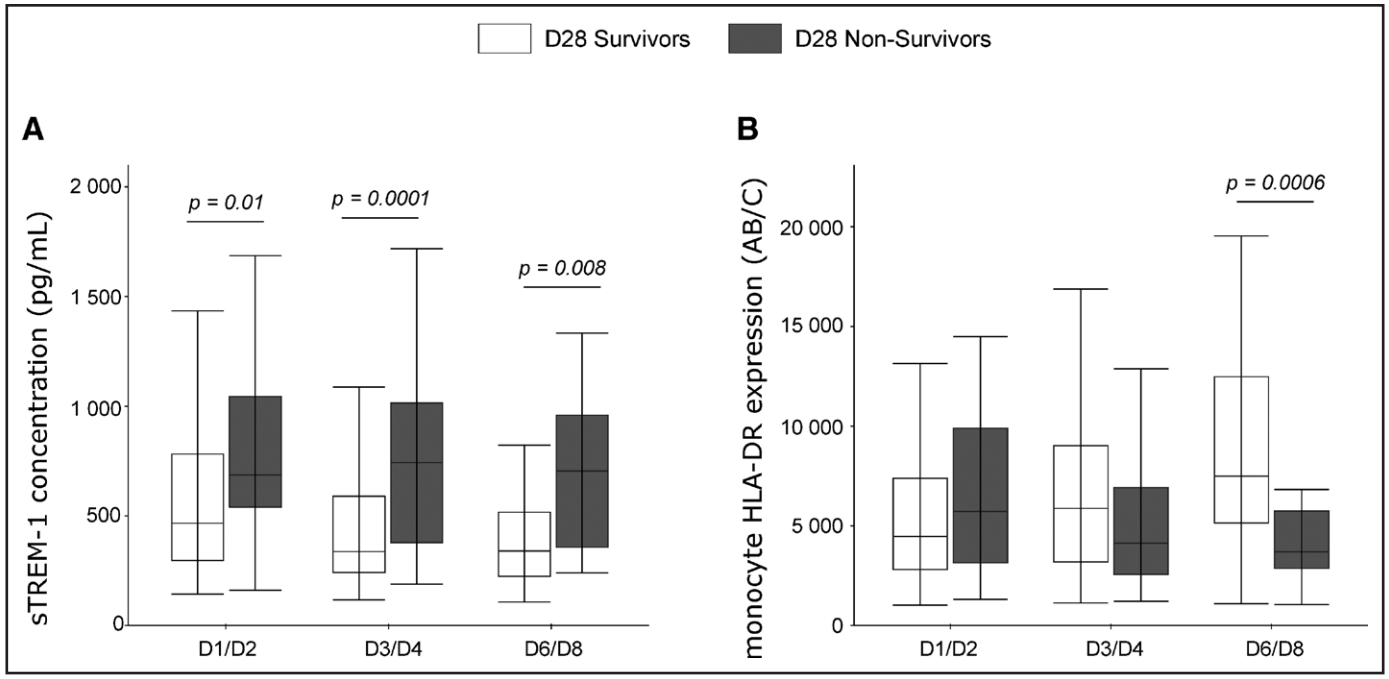

Figure 1. Soluble form of triggering receptor expressed on myeloid cells 1 (sTREM-1) concentration and human leucocyte antigen-DR on monocytes (mHLA-DR) expression in survivors and nonsurvivors septic shock patients. **A**, sTREM-1 concentrations were measured by enzyme-linked immunosorbent assay in survivors (*white boxes*, $n = 87$) and nonsurvivors septic patients (*gray boxes*, $n = 29$) at D1/D2, D3/D4, and D6/D8. Results were expressed as pg/mL. **B**, HLA-DR expression on monocytes was measured in survivors and nonsurvivors septic patients and expressed as number of anti-HLA-DR antibodies bound per cell (AB/C). Results are presented as Tukey box plots. Mann-Whitney tests were used for comparisons between groups at each time point. Only *p* values below 0.05 are shown.

D6/D8 remained independently associated with the occurrence of nosocomial infection (sHR [95% CI], 3.61 [1.39–9.34]; $p = 0.008$). Regarding sTREM-1, in univariate analysis, this parameter was not significantly different between patients who presented or not secondary infections at any given time point, although the analysis was close to the limit of significance at D6/D8 ($p = 0.05$).

Combined Analysis of sTREM-1/mHLA-DR Levels at D6/D8 and Association With Nosocomial Infections

Finally, to assess the potential of the combination of sTREM-1 and mHLA-DR in regard with association with risk of nosocomial infections, we defined subgroups of patients according to high or low biomarker values at D6/D8 (defined by median values). In that respect, patients with high sTREM-1 (above median, i.e., >392 pg/mL) and low mHLA-DR (below median, i.e., less than 6668 AB/C) at this time point were classified into an EP group (thereafter called EP group) and were compared with all other patients. **Table 2** lists clinical characteristics of EP patients in comparison with the rest of the cohort. Initial SOFA score was significantly increased in EP subgroup ($p = 0.004$) as well as lactate concentration at diagnosis of shock ($p = 0.02$) and plasma creatinine concentration ($p < 0.01$). Cumulative incidence curves of nosocomial infections in EP subgroup and other patients are presented in **Figure 3*B***. EP patients presented higher rate of nosocomial infection (60 %) compared with other patients within the first 28 days after ICU admission (15.7 %). In univariate analysis, this difference was significant ($p < 0.001$). Importantly, the multivariable analysis confirmed that combined marker evaluation was independently associated with a higher risk of nosocomial infections (sHR [95% CI], 4.65 [1.98–10.9]; $p < 0.001$). This association was stronger than observed with mHLA-DR expression alone (i.e., sHR = 3.61). In this combined analysis, neither age nor the initial SOFA score was risk factors.

DISCUSSION

TREM-1 is an innate immunity receptor expressed on myeloid cells such as monocytes and neutrophils

TABLE 1.
Baseline Demographic and Clinical Characteristics of Patients With or Without Secondary Infections

Clinical and Biological Parameters	No nosocomial infection at day 28 ($n = 85$)	Nosocomial infection at day 28 ($n = 25$)	p
General characteristics			
Gender (male)	52 (61.2)	14 (56.0)	0.816
Age	71.3 (62.0–78.9)	69.1 (64.5–77.3)	0.620
Body mass index (kg/m^2)	24.97 (21.89–30.72)	27.68 (22.86–29.41)	0.404
McCabe score			
0	41 (49.4)	15 (60.0)	0.631
1	35 (42.2)	8 (32.0)	
2	7 (8.4)	2 (8.0)	
Severity scores at admission			
Sepsis-related Organ Failure Assessment score	9.0 (7.0–11.0)	9.0 (8.0–11.0)	0.719
Charlson score	2.0 (0.0–4.0)	2.0 (1.0–3.0)	0.786
Simplified Acute Physiology Score II score	56.5 (47.8–73.3)	60.5 (50.8–72.8)	0.665
Pao_2/Fio_2 (mm Hg)	200.0 (161.0–278.0)	200.0 (137.3–277.3)	0.605
Primary site of infection			
Intra-abdominal	42 (49.4)	9 (36.0)	0.003
Pulmonary	9 (10.6)	10 (40.0)	
Other	34 (40.0)	6 (24)	
Context			
Medical	18 (21.7)	9 (37.5)	0.239
Surgery	65 (78.3)	15 (62.5)	
Major surgery	13 (15.7)	1 (4.2)	
Pancreatitis	1 (1.2)	1 (4.2)	
Biological values in plasma at D1			
pH	7.38 (7.28–7.44)	7.34 (7.29–7.38)	0.167
Arterial lactate (mmol/L)	2.60 (1.90–3.63)	2.50 (1.70–3.80)	0.762
Leucocyte count (G/L)	15.30 (9.17–26.60)	16.90 (12.35–21.27)	0.645
Neutrophil count (G/L)	13.70 (8.37–24.00)	13.82 (11.15–17.05)	0.976
Lymphocyte count (G/L)	1.08 (0.64–1.70)	0.93 (0.60–1.49)	0.584
Monocyte count (G/L)	0.60 (0.41–1.16)	0.80 (0.52–1.10)	0.395
Hemoglobin (g/dL)	10.35 (8.80–11.80)	9.90 (9.10–12.00)	0.943
Platelet count (G/l)	205.50 (129.25–320.25)	264.50 (171.00–392.25)	0.058
Creatinine (µmol/l)	134.00 (90.00–222.00)	171.00 (86.00–242.00)	0.663
Exposure to invasive devices			
Urinary catheter	78 (91.8)	24 (96.0)	0.780
Central venous catheter	82 (96.5)	25 (100.0)	0.800
Orotracheal intubation	66 (77.6)	23 (92.0)	0.188
Adjuvant therapy			
Hydrocortisone	34 (40.0)	12 (48.0)	0.630

Results are shown as medians and interquartile ranges (Q1–Q3) for continuous variables or numbers and percentages for categorical variables. Patients were separated in two groups based on the occurrence of nosocomial infection within the first 28 d after ICU admission. Sepsis-related Organ Failure Assessment and Simplified Acute Physiology Score II scores were calculated during the first 24 hr after ICU admission. Hepatic resection, pancreatic and esophageal surgeries, cystectomy with Bricker reconstruction, and cardiac surgery were considered as major surgery. Data were compared using Fisher exact test or χ^2 for categorical data and the Wilcoxon test for continuous data.

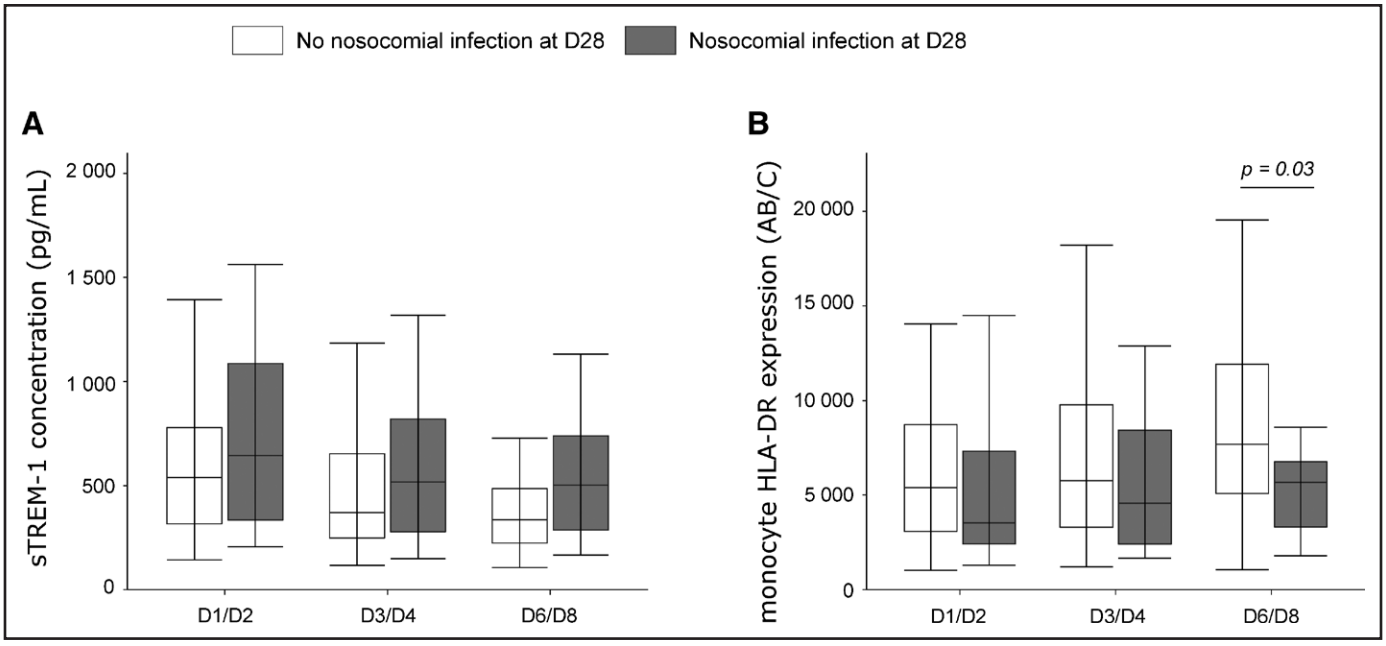

Figure 2. Soluble form of triggering receptor expressed on myeloid cells 1 (sTREM-1) concentration and human leucocyte antigen-DR on monocytes (mHLA-DR) expression in septic shock patients with or without nosocomial infection. **A**, sTREM-1 concentrations were measured by enzyme-linked immunosorbent assay in patients with secondary infection (*gray boxes*, $n = 25$) and in patients with no secondary infections (*white boxes*, $n = 85$) at D1/D2, D3/D4, and D6/D8. Results were expressed as pg/mL. **B**, HLA-DR expression on monocytes was measured in septic patients with or without secondary infections and expressed as number of anti-HLA-DR antibodies bound per monocyte (AB/C). Results are presented as Tukey box plots. Mann-Whitney tests were used for comparisons between groups at each time point. Only *p* values below 0.05 are shown.

involved in their activation and induction of effector functions (cytokine synthesis, degranulation, oxidative burst, and antigen presentation) (19–21). Once activated, sTREM-1 is released by immune cells due to surface proteolysis and can be considered as a marker of TREM-1 pathway activation (26).

Described as a marker of inflammation in sepsis, sTREM-1 has been largely studied as a diagnostic marker of infection, in blood or bronchoalveolar lavage (28, 29, 36). In addition, its association with mortality was repeatedly investigated, and elevated sTREM-1 concentrations at ICU admission were associated with higher mortality in sepsis (30–33, 36, 37). We confirmed these results in the current study as we observed that nonsurvivors presented with higher sTREM-1 levels at D1/D2 than survivors. We also provide additional information by showing that this association persisted during the first week after admission. Indeed, we observed persistently higher plasma sTREM-1 levels at D6/D8 in nonsurvivors compared with survivors. Few studies investigated the longitudinal evolution of sTREM-1 concentrations in large cohorts of septic patients and showed the persistence of high plasma sTREM-1 in some patients in association with mortality. In addition, few inflammatory markers present with such kinetic overtime and persistent association with mortality over the first week after admission. For example, although increased procalcitonin and C-reactive protein concentrations at ICU admission have repeatedly been shown to be associated with higher mortality, no association was found when these markers were measured at later time points (36, 37). IL-6, widely studied in septic shock with high concentration at admission, provided heterogeneous results regarding its mortality predictive value over time (37–39).

Considering that sTREM-1 is released from membrane as a marker of TREM-1 pathway activation and since sTREM-1 displays a short half-life in vivo (26), continuous elevated levels in nonsurvivors may suggest the persistence in some patients of continuous TREM-1 pathway stimulation and cellular activation. This could be explained by the incomplete or the absence of clearance of initial infectious foci in some patients with sepsis. In line, in an autopsy study of patients who died of septic shock, 80% had uncontrolled septic outbreaks at time of death, which corroborates with the presence of a high sTREM-1 (40).

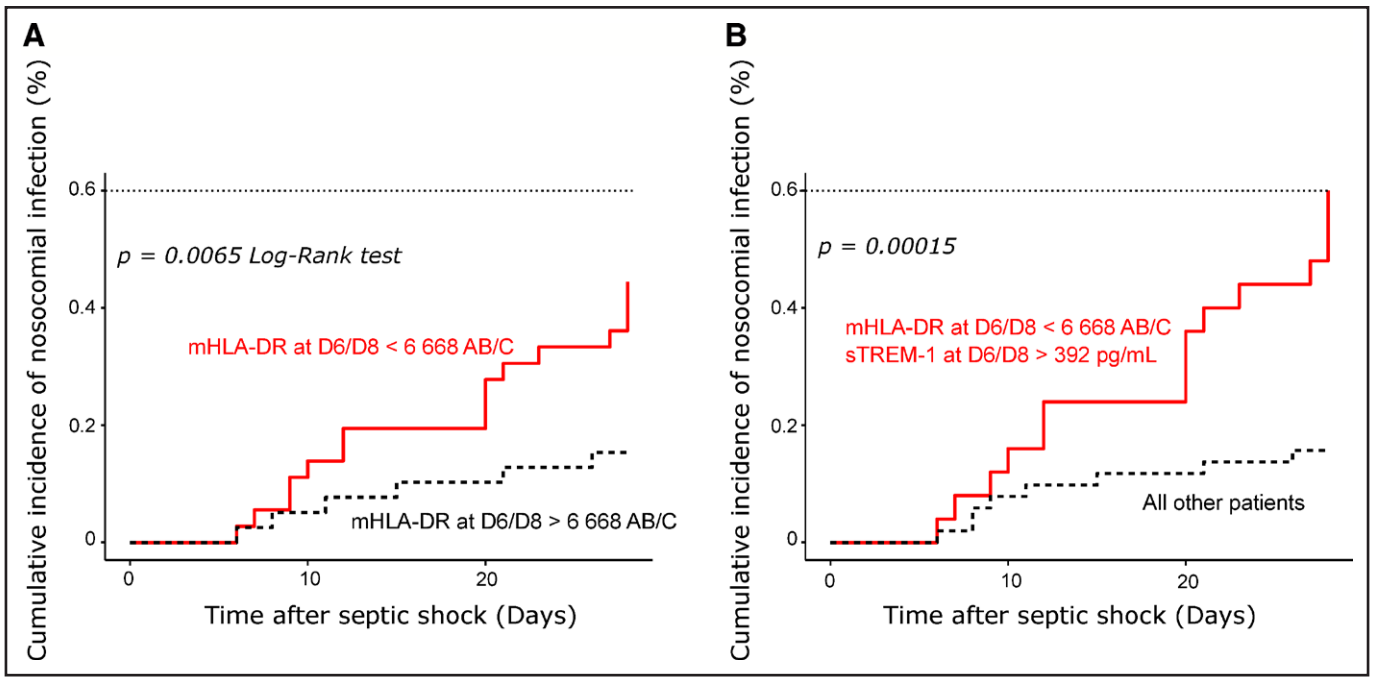

Figure 3. Cumulative incidence curves of nosocomial infection occurrence in septic shock patients stratified based on human leucocyte antigen-DR on monocytes (mHLA-DR) and/or soluble form of triggering receptor expressed on myeloid cells 1 (sTREM-1) values at D6/D8. **A**, Septic shock patients were stratified based on median mHLA-DR value measured at D6/D8 (i.e., 6668 antibodies bound per monocyte [AB/C]). The occurrence of nosocomial infections was monitored in both groups during 28 d after ICU admission. *Red full line* represents the group of patients with mHLA-DR at D6/D8 below median value. *Black dotted line* represents the group of patients with mHLA-DR value above median. Cumulative incidence curves were assessed using a Fine and Gray competing risk model. **B**, Septic shock patients were stratified based on median mHLA-DR value (i.e., 6668 AB/C) and median sTREM-1 value (i.e., 392 pg/mL) measured at D6/D8. The occurrence of nosocomial infections during 28 d after ICU admission was monitored in patients with both mHLA-DR below median value and sTREM-1 above median value at D6/D8 (*red full line*) and in the rest of the cohort (*black dotted line*). Cumulative incidence curves were assessed using a Fine and Gray competing risk model.

Importantly, the persistence of such low grade inflammation and cellular activation in some patients has been proposed as one mechanism sustaining the process of sepsis-induced immunosuppression (41, 42). Yet, to our knowledge, no study has investigated the prognostic value of sTREM-1 regarding the risk of occurrence of nosocomial infection in adult septic shock patients, whereas sepsis-induced immunosuppression is known to contribute to increased nosocomial infections risk. Only two studies evaluated its association with the occurrence of nosocomial infections, and both were restricted to the pediatric context (ventilator-associated pneumonia in neonates and catheter infections in a pediatric population with severe digestive disorders [43, 44]). In addition, no study has investigated sTREM-1 along with usual markers of sepsis-induced immunosuppression. The main interest of the present study is, thus, to provide for the first time both sTREM-1 values and mHLA-DR expression in a unique cohort of septic shock patients.

As expected, we report that delayed low mHLA-DR at D6/D8 was independently associated with nosocomial infections (sHR = 3.61). At this time point, although sTREM-1 concentration was not significantly different between patients who developed or not secondary infections, we showed that patients with persistently high sTREM-1 and low mHLA-DR presented with a high risk of nosocomial infection independently of usual confounding factors such as older age or initial clinical severity. Importantly, the combination of high sTREM-1 value with results of low mHLA-DR improved the strength of the association with infection compared with results obtained with mHLA-DR alone with an sHR of 4.65 versus 3.61 for mHLA-DR alone.

These results are in line with recent biomarker development, which combined pro and anti-inflammatory markers to improve prediction of deleterious outcomes such as secondary infections (45, 46). This was, for example, described in trauma patients with the association of plasma IL-6 levels and mHLA-DR (47).

TABLE 2.
Baseline Demographic and Clinical Characteristics of the Extreme Phenotype Group vs the Rest of the Cohort

Clinical and Biological Parameters	Extreme Phenotype Subgroup ($n = 25$)	Rest of Cohort ($n = 51$)	p
General characteristics			
Gender (male)	13 (52.0)	36 (70.6)	0.182
Age	72.1 (54.5–79.9)	68.8 (61.6–77.4)	0.214
Body mass index (kg/m^2)	26.23 (21.48–29.41)	25.40 (21.30–29.41)	0.706
McCabe score			
0	13 (54.2)	23 (46.0)	0.686
1	9 (37.5)	24 (48.0)	
2	2 (8.3)	3 (6.0)	
Severity score			
Sepsis-related Organ Failure Assessment score	11.0 (9.0–13.0)	9.0 (8.0–10.0)	0.004
Charlson score	2.0 (1.0–3.0)	2.0 (1.0–4.0)	0.512
Simplified Acute Physiology Score II score	65.5 (55.3–76.0)	56.5 (48.8–69.0)	0.138
Pa_{O_2}/F_{IO_2} at admission (mmHg)	163.0 (111.0–218.5)	201.0 (166.0–259.0)	0.102
Primary site of infection			
Intra-abdominal	11 (44.0)	28 (54.9)	0.430
Pulmonary	7 (28.0)	8 (15.7)	
Other	7 (28.0)	15 (29.4)	
Context			
Medical	6 (26.1)	14 (28.0)	
Surgery	17 (73.9)	36 (72.0)	0.772
Major surgery	2 (8.7)	7 (14.0)	
Pancreatitis context	1 (4.3)	1 (2.0)	
Biological values in plasma at D1			
pH	7.31 (7.28–7.40)	7.39 (7.31–7.43)	0.051
Arterial lactate (mmol/l)	2.80 (2.10–4.65)	2.20 (1.60–2.87)	0.020
Leucocyte count (G/l)	16.60 (6.25–26.54)	15.01 (9.95–24.04)	0.631
Neutrophil count (G/l)	12.80 (6.90–20.25)	12.60 (9.09–20.50)	0.613
Lymphocyte count (G/l)	0.96 (0.48–1.65)	1.10 (0.71–1.77)	0.304
Monocyte count (G/l)	0.54 (0.22–0.85)	0.70 (0.50–1.30)	0.083
Hemoglobin (g/dL)	10.70 (9.20–12.70)	10.50 (9.30–11.80)	0.619
Platelet count (G/l)	188.00 (138.00–274.25)	225.50 (143.50–326.50)	0.564
Creatinine (µmol/l)	230.00 (183.00–281.00)	126.00 (82.50–185.00)	< 0.001
Exposure to invasive devices			
Urinary catheter	24 (96.0)	45 (88.2)	0.498
Central venous catheter	25 (100.0)	25 (100.0)	
Orotracheal intubation	22 (88.0)	43 (84.3)	0.935
Adjuvant therapy			
Hydrocortisone	15 (60.0)	16 (31.4)	0.033

Results are shown as medians and interquartile ranges (Q1–Q3) for continuous variables or numbers and percentages for categorical variables. The extreme phenotype subgroup is composed of patients having plasma concentration of soluble form of triggering receptor expressed on myeloid cells 1 above the median and human leucocyte antigen-DR on monocytes below the median at D6/D8. Sepsis-related Organ Failure Assessment and Simplified Acute Physiology Score II scores were calculated during the first 24 hr after ICU admission. Hepatic resection, pancreatic and esophageal surgeries, cystectomy with Bricker reconstruction, and cardiac surgery were considered as major surgery. Data were compared using Fisher exact test or χ^2 for categorical data and the Wilcoxon test for continuous data.

The combination of several markers of immune response is currently proposed to better understand/capture the complex pathophysiology of sepsis and the heterogeneity of patients' immune response evolving overtime (48). Although we cannot elaborate on the causative relationship between both markers as this was not within the scope of this study, this result supports the hypothesis stating that persistence of a low grade inflammation belongs to mechanisms sustaining the process of sepsis-induced immunosuppression, which may then amplify the risk for nosocomial infection (41).

This study presents some limitations. First due to the retrospective selection of patients with available plasma samples for sTREM-1 measurement, TREM-1 expression on circulating leucocytes and TREM-1 mRNA levels were not evaluated. Second, these results need to be validated in an external prospective cohort including a larger number of patients to confirm the association with outcomes. Finally, the functional role of persistent sTREM-1 in the pathophysiology of sepsis needs to be evaluated in a dedicated study including, for example, ex vitro studies with cells from patients incubated with recombinant sTREM-1.

CONCLUSIONS

In addition to having a prognosis interest on mortality, sTREM-1, when combined with mHLA-DR, may help to better identify immunosuppressed patients at risk of nosocomial infections. Upon confirmation in a dedicated prospective study, this biomarker combination may help to identify patients who might benefit from immune-adjuvant therapy.

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