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Low levels of CD72 and CD100 expression on circulating lymphocytes in immunosuppressive phase of sepsis is associated with mortality in septic patients

Yang Yang, Jiajun Chen, Manli Tang, Chengla Yi, Wei Gao, Xiangjun Bai*, Zhanfei Li and Fan Yang*

Abstract

Background: Despite improvements in antimicrobial therapy and supportive care, sepsis is still a major public health issue. Recently, CD100 and its receptor in the immune system CD72 were shown to play a major role in immune regulation. The purpose of this study was to investigate the expression and clinical correlations of CD72 and CD100 on circulating lymphocytes of septic patients.

Methods: In total, 24 healthy controls and 54 septic patients were enrolled in this study. Considering the focus of the current study was on the immunosuppressive phase of sepsis, blood samples of patients were collected at days 3–4 after the onset of sepsis. The levels of CD72 and CD100 expression on circulating lymphocytes were measured by flow cytometry and serum IL-6, IL-10, and immunoglobulin M levels were determined by enzyme-linked immunosorbent assay.

Results: Our results showed that the levels of CD100 expression on T cells and CD72 expression on B cells were significantly lower in septic patients. Similarly, a significant decrease in the expression levels of CD72 and CD100 was observed in non-survivors compared with survivors. In addition, the reduction of immunoglobulin M levels and lymphocyte counts were correlated with the low CD72 and CD100 expression levels. Multivariate logistic regression analysis showed that the percentage of CD100⁺/CD8⁺ T cells and CD72⁺/CD19⁺ B cells were independent predictors of 28-day mortality in septic patients. Simultaneously, the receiver operating characteristic curve analysis showed that the combination of the percentage of CD100⁺/CD8⁺ T cells and sequential organ failure assessment score had the best predictive value of mortality risk.

Conclusions: Our study demonstrated that the decrease of the levels of CD72 and CD100 expression on circulating lymphocytes after 3–4 days of sepsis had a close correlation of the 28-day mortality of septic patients. Thus, CD72 and CD100 are promising biomarkers for assessing the prognosis of patients with sepsis.

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Trial registration: Peripheral blood lymphocytes analysis detects CD72 and CD100 alteration in trauma patients; ChiCTR1900026367; Registered 4 October 2019; <http://www.chictr.org.cn>.

Keywords: Sepsis, Lymphocyte, CD72, CD100, Mortality, Immunosuppressive

Background

Sepsis is defined as life-threatening organ dysfunction caused by a dysregulated host response to infection [1]. Despite improvements in antimicrobial therapy and supportive care, sepsis is still a major public health issue. To date, existing epidemiologic studies suggest that global estimates of 31.5 million sepsis and 19.4 million severe sepsis cases, with potentially 5.3 million deaths annually [2]. In sepsis, the immune response that is initiated by an invading pathogen fails to return to homeostasis, thus culminating in a pathological syndrome that is characterized by sustained excessive inflammation and immune suppression [3]. Moreover, a number of studies have demonstrated that severe immunosuppression was the key cause of high mortality [4]. Therefore, knowledge of the complex immunopathological mechanisms of sepsis-induced immunosuppression is essential for precision medicine in sepsis.

CD100 (also called SEMA4D) is a 150-kDa protein that belongs to the class IV of the semaphorin family [5]. Generally, CD100 is expressed abundantly on the surface of T cells and weakly on dendritic cells (DCs), B cells, and NK cells [5–8]. Both in human and mice it was demonstrated that CD100 influenced T-cell activation, DCs activation and migration, B-cell survival, NK-cell killing activity [5–8]. CD100 functions as a ligand and binds to three different receptors: the low-affinity receptor CD72, in lymphoid tissue, and the high-affinity receptors plexin B1 and plexin B2, in non-lymphoid tissue [9–11]. CD72 (originally Lyb-2) is a 45-kDa type II transmembrane glycoprotein that belongs to the calcium-dependent C-type lectin family [12, 13]. CD72 is expressed primarily on B cells and weakly on T cells, and it is known as a negative regulator of B cells [14]. Furthermore, previous studies have highlighted that the interaction of CD100 and CD72 is essential for maintaining immunological homeostasis [15, 16].

Although CD100 and CD72 have attracted particular attention due to their extensive functions in the immune system, research has mainly focused on the roles of CD72 and CD100 in viral infections and autoimmune diseases [17–19], little research has explored the relationship between CD72/CD100-related molecules and the progress of sepsis, and evaluated which CD72/CD100-related molecules are reliable biomarkers to predict mortality in the immunosuppressive phase of sepsis. Therefore, we designed this study to investigate whether CD72 and CD100 expression on circulating lymphocytes

during the immunosuppressive phase of septic patients can be used to evaluate the prognosis of septic patients. Furthermore, we evaluated the prognostic value of the combination of the best CD72/CD100-related predictors and conventional clinical risk parameters in septic patients.

Materials and methods

Patients and controls

Fifty-four septic patients (included twenty-two septic shock patients) were enrolled on admission to the Department of Emergency and Traumatic Surgery of Tongji Hospital of Tongji Medical College of Huazhong University of Science and Technology at days 1–2 after sepsis was diagnosed. According to the Sepsis-3 [1], sepsis was defined as life-threatening organ dysfunction caused by a dysregulated host response to infection. And organ dysfunction was identified as an acute change in total sequential organ failure assessment (SOFA) score ≥ 2 points consequent to the infection. Moreover, patients with septic shock can be identified with a clinical construct of sepsis with persisting hypotension requiring vasopressors to maintain mean arterial pressure (MAP) ≥ 65 mmHg and having a serum lactate level > 2 mmol/L (18 mg/dL) despite adequate volume resuscitation. Besides, patients who met the following criteria were excluded: (a) age less than 18 years; (b) patients with pregnancy; (c) sepsis occurred more than 3 days prior to admission; (d) patients who died within 2 days after the onset of sepsis; (e) use of immunosuppressive medication in the past 8 weeks; (f) patients with malignancy, autoimmune diseases or chronic viral infections (hepatitis, HIV). In this study, blood samples of septic patients were collected at days 3–4 after the onset of sepsis due to the investigation's focus on the immunosuppressive phase of sepsis [20, 21]. In addition, twenty-four age- and sex-matched healthy adult volunteers without medical problems were recruited as healthy controls. This protocol was approved by the medical ethics committee of Tongji Hospital affiliated to Tongji Medical College of Huazhong University of Science and Technology, and informed consent was obtained from all participants according to the Declaration of Helsinki.

Data collection

The clinical data, including age, gender, the counts of white blood cell (WBC), the counts of lymphocyte (LC), the levels of C-reaction protein (CRP), site of infection,

microbial data, SOFA score, simplified acute physiology score II (SAPS II) and acute physiology and chronic health evaluation II (APACHE II) score were recorded at days 3–4 after the onset of sepsis. In addition, the mortality during the 28-day study period was also recorded.

Flow cytometry

Blood samples of septic patients were collected in EDTA vacutainer and transported to the laboratory at 4 °C within 2 h. Erythrocytes were lysed using Lysing Buffer (BD Bioscience) and cells were stained with fluorescence-conjugated antibodies (CD3, CD4, CD8, CD19, CD72, CD100 by BD Bioscience). According to the manufacturer's recommendations antibodies were used: BV510-labeled anti-CD3 (5 µl, clone HIT3a), BB700-labeled anti-CD4 (5 µl, clone SK3), PECy7-labeled anti-CD8 (5 µl, clone RPA-T8), PE-labeled anti-CD19 (20 µl, clone HIB19), FITC-labeled anti-CD72 (20 µl, clone J4-117), and Alexa647-labeled anti-CD100 (5 µl, clone A8) per 100 µl of whole blood. After 30 min of light-protected incubation, the cells were washed, centrifuged, and subjected to flow-cytometric analysis. Samples were run on a CytoFlex Flow cytometer (Beckman Coulter, Inc.) and analyzed using CytoFlex Software version 2.0 (Beckman Coulter, Inc.). Lymphocytes were gated by side scatter (SSC) and forward scatter (FSC). T cells were defined as CD3⁺ (subsequent analysis for CD4⁺ and CD8⁺), B cells were defined as CD19⁺ (Fig. 1). Results were expressed as percentages.

Cytokine and immunoglobulin M measurement

Serum levels of interleukin-6 (IL-6) and IL-10 were determined using commercially available human ELISA kits (ELK Biotechnology Co., Ltd, Wuhan, China) according to the manufacturer's instructions. In addition, serum levels of immunoglobulin M (IgM) were tested by human IgM ELISA kit (BD Biosciences).

Statistical analysis

Considering that the skewed distribution of most parameters, data were expressed as median (interquartile ranges), frequencies and percentages unless otherwise indicated. Comparisons between groups were performed using Pearson χ^2 test or Fisher exact test for categorical variables, as appropriate. For continuous data, the Shapiro-Wilk test was used for testing normality. Normally distributed data were compared using Student's *t* test or ANOVA for two or more than two groups, respectively. Mann-Whitney *U* test or Kruskal-Wallis test was employed for data of non-normal distribution. To determine the discriminative power of CD72 and CD100 expression levels in septic patients for 28-day mortality, the receiver operating characteristic (ROC) curves were constructed, and the significance of the differences between AUCs was estimated by using DeLong test.

Spearman's rank correlation coefficient was used to test correlations between different parameters. For septic survival studies, Kaplan-Meier analyses were performed, and log-rank test was applied for the comparisons of survival distributions. To identify which CD100/CD72 related molecules were independently associated with 28-day mortality in septic patients, univariate and multivariate logistic regression analysis were performed. Statistical analyses and graphics were done using SPSS 23.0 (IBM Corp., Armonk, NY, USA), GraphPad Prism 8.02 (GraphPad Software, Inc., La Jolla, CA, USA) and MedCalc 12.7 (Ostend, Belgium). All statistical tests were two-tailed, and *P* < 0.05 were considered statistically significant.

Results

Patient characteristics

The demographic and clinical characteristics of the patients and healthy controls are shown in Tables 1, 2, and S1. In the patient group, higher SOFA score, APACHE II score and SAPS II indicated a high level of severity. Meanwhile, the comparison of survivors and non-survivors also showed significant differences in SOFA score, APACHE II score, and SAPS II. Moreover, the major site of infection was thorax and the major cause of sepsis was bacterial infection (gram-positive).

The expression levels of CD72 and CD100 were both decreased in septic patients

The expression of CD72 and CD100 were measured on circulating CD4⁺ T cells, CD8⁺ T cells, and CD19⁺ B cells at days 3–4 after the onset of sepsis (Fig. 2). In this study, the levels of CD72 expression on T cells and CD100 expression on B cells were very low both in healthy controls and patients (data not shown), so the comparisons between them were not performed. Comparison with healthy controls, the percentages of circulating CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, and CD72⁺/CD19⁺ B cells were significantly lower in septic patients (Table 1 and Fig. 2). Moreover, they were also obviously different between septic patients and septic shock patients (Table 1). Meanwhile, in non-survivors, the percentages of CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, and CD72⁺/CD19⁺ B cells were remarkably lower in comparison with survivors (Table 2 and Fig. 2).

The alteration of serum cytokine and IgM levels were related to the expression levels of CD72 and CD100

High levels of pro- and anti-inflammatory cytokines were common in septic patients. In this study, we showed serum IL-6 and IL-10 levels were significantly elevated in septic patients (Table 1). However, only serum IL-6 levels were dramatically increased in septic shock patients compared to septic patients (Table 1). Similar results were also

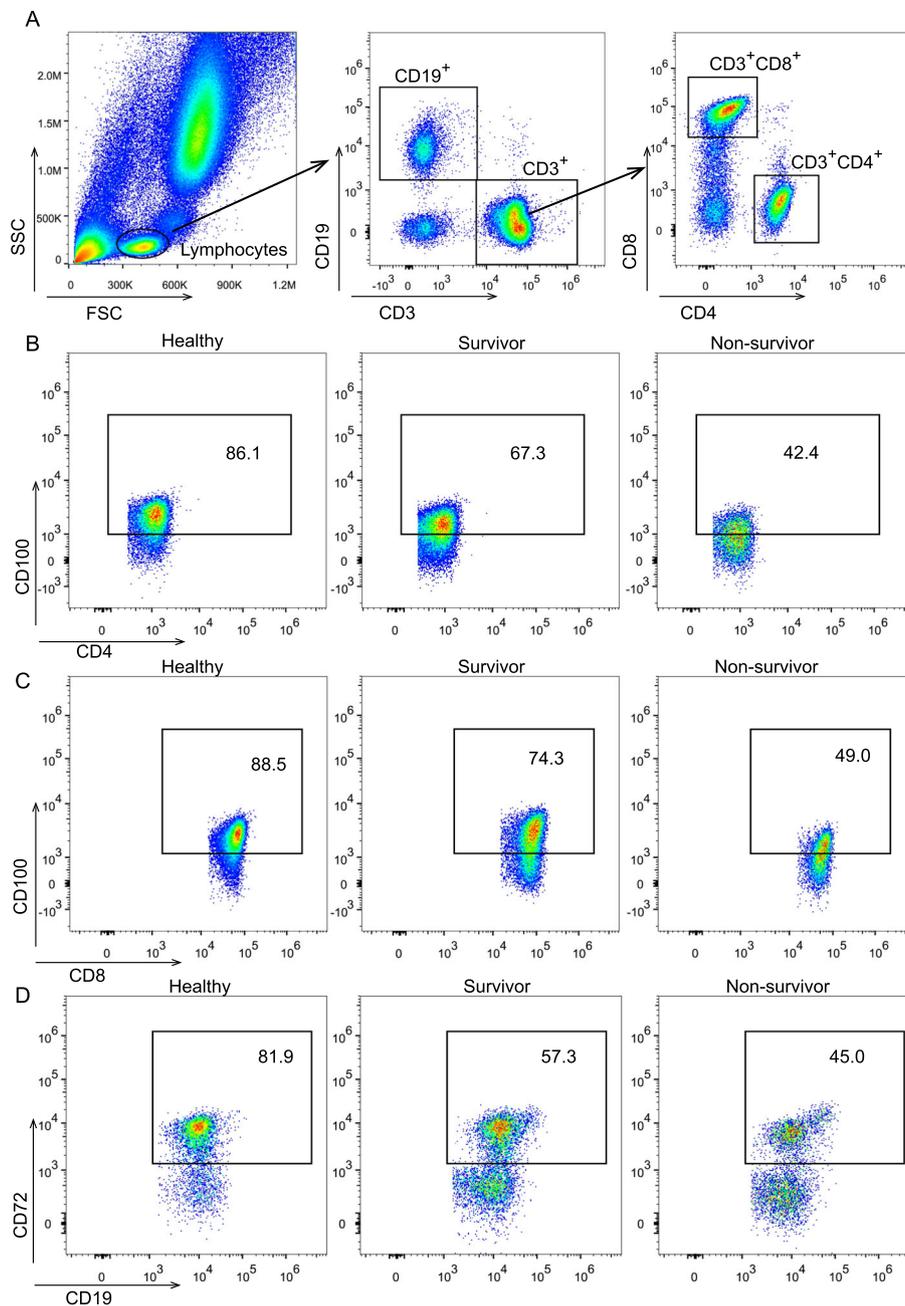


Fig. 1 Representative flow pseudo-color of lymphocytes gating strategy (a) and the levels of CD100 expression on CD4⁺ T cells (b), CD100 expression on CD8⁺ T cells (c), CD72 expression on CD19⁺ B cells (d), in healthy controls (n = 24), septic survivors (n = 38), and septic non-survivors (n = 16), respectively

observed when compared non-survivors with survivors (Table 2). Additionally, serum IL-6 levels were reversely correlated with the percentages of CD100⁺/CD4⁺ T cells and the percentages of CD100⁺/CD8⁺ T cells ($r = -0.3541$ and -0.3124 , respectively) (Fig. 3b and f). On the other hand, serum IgM levels dropped in turn in healthy controls, survivors, and non-survivors (Tables 1, 2). Meanwhile, significant positive correlations were observed between serum IgM levels and the percentages of

CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, and CD72⁺/CD19⁺ B cells ($r = 0.3031$, 0.5168 , and 0.324 , respectively) (Fig. 3d, h, and l).

Lymphopenia was correlated with the low expression levels of CD72 and CD100

Lymphopenia was ubiquitous in septic patients (Table 1), including CD4⁺ T cells, CD8⁺ T cells and CD19⁺ B cells (Table S2), and more significant reduction was

Table 1 Baseline characteristics of healthy controls and patients

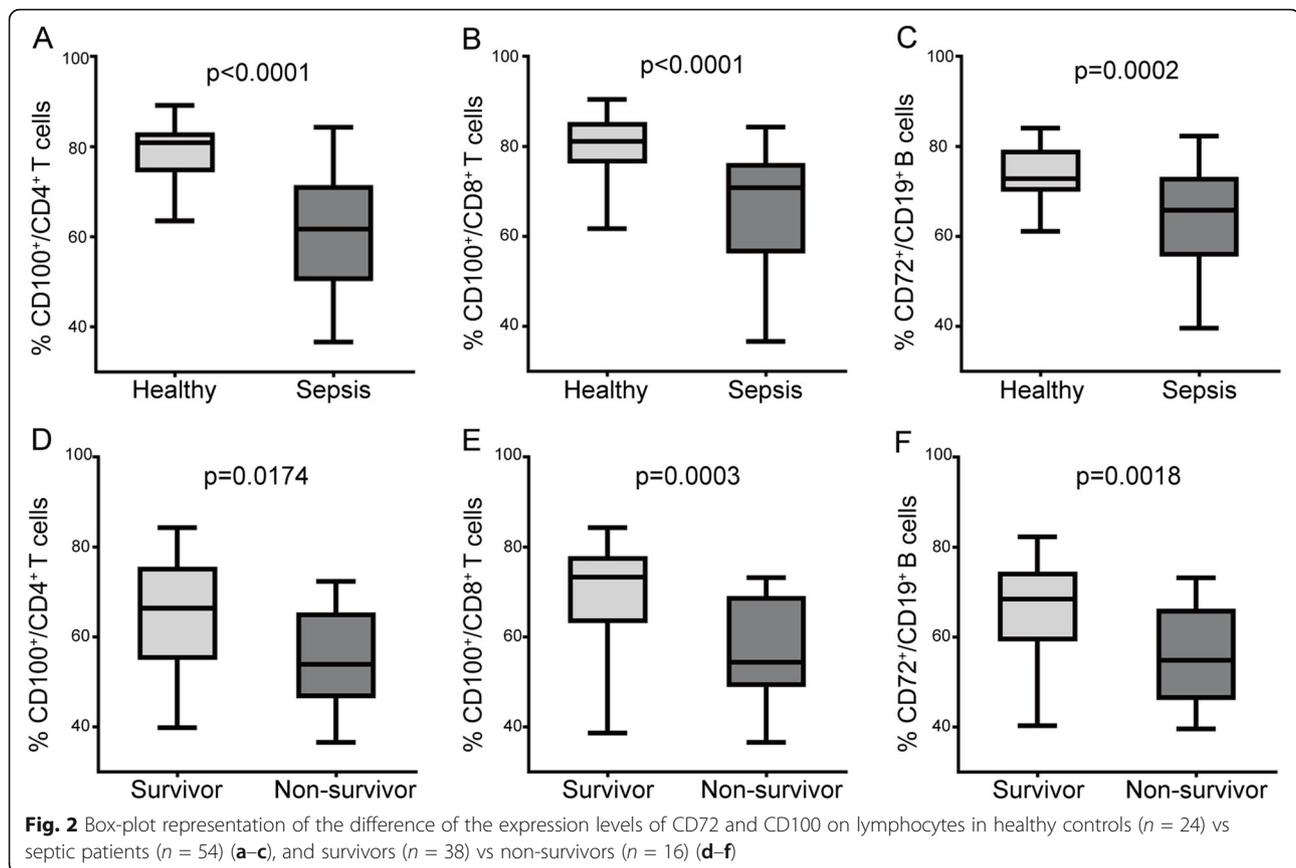
Parameters	Healthy	Sepsis	Septic shock	P
Number	24	32	22	NA
Age (years)	47 (44–52)	51 (47–58)	47 (42–53) ^b	0.0474
Male, n (%)	17 (70.8%)	27 (84.4%)	16 (72.7%) ^b	0.423
WBC (10 ⁹ /L)	7.58 (7.34–7.88)	14.22 (12.68–16.57)	14.30 (13.22–15.77) ^b	<0.0001
LC (10 ⁹ /L)	2.05 (1.90–2.15)	0.91 (0.77–1.09)	0.77 (0.68–0.92) ^a	<0.0001
SOFA score	NA	7 (5–10)	9 (7–11) ^a	0.047
SAPS II	NA	29 (23–34)	36 (26–46) ^a	0.0311
APACHE II score	NA	14 (12–17)	17 (15–21) ^a	0.008
CRP (mg/L)	1.3 (0.8–1.8)	126.7 (104.2–146.5)	131.9 (107.8–168.7) ^b	<0.0001
IgM (g/L)	1.78 (1.48–2.15)	0.84 (0.76–1.0)	0.83 (0.72–0.93) ^b	<0.0001
IL-6 (pg/ml)	10.6 (9.65–11.6)	80.0 (64.81–91.54)	88.58 (78.58–95.49) ^a	<0.0001
IL-10 (pg/ml)	10.09 (9.14–11.17)	87.6 (75.88–100.9)	88.43 (72.94–98.84) ^b	<0.0001
CD100 ⁺ /CD4 ⁺ T cells %	80.9 (74.3–83.2)	67.0 (58.7–73.3)	52.8 (48.3–63.2) ^a	<0.0001
CD100 ⁺ /CD8 ⁺ T cells %	81.2 (76.2–85.5)	71.5 (62.7–77.4)	59.8 (48.0–75.4) ^a	<0.0001
CD72 ⁺ /CD19 ⁺ B cells %	72.8 (69.9–79.3)	67.9 (59.8–73.9)	58.5 (48.3–72.4) ^a	<0.0001
28-day mortality, n (%)	NA	7 (21.9%)	9 (40.9%) ^b	0.132

Data are shown as median (interquartile ranges), frequencies and percentages. ANOVA and Kruskal-Wallis tests were performed for multi-group comparisons. ^aDenotes $P < .05$ and ^bdenotes $P > .05$ comparing sepsis group and septic shock group. NA Not applicable, WBC Counts of white blood cell, LC Counts of lymphocyte, SOFA Sequential organ failure assessment, SAPS II Simplified acute physiology score II, APACHE II Acute physiology and chronic health evaluation II, IgM Immunoglobulin M, CRP C-reaction protein, IL Interleukin

Table 2 Baseline characteristics of septic survivors and non-survivors

Parameters	Survivor	Non-survivor	Overall population	P
Number	38	16	54	NA
Age (years)	49 (45–56)	50 (45–61)	49 (45–56)	0.458
Male, n (%)	31 (81.6%)	12 (75%)	43 (79.6%)	0.714
WBC (10 ⁹ /L)	14.22 (12.43–15.77)	14.85 (13.68–17.45)	14.23 (13.09–16.44)	0.0709
LC (10 ⁹ /L)	0.89 (0.75–1.05)	0.77 (0.66–0.90)	0.86 (0.71–1.03)	0.0316
SOFA score	7 (6–9)	10 (9–13)	8 (6–10)	<0.0001
SAPS II	28 (24–34)	43 (31–50)	31 (25–42)	0.0011
APACHE II score	14 (12–16)	19 (16–22)	15 (13–19)	0.0005
CRP (mg/L)	124.5 (101.1–148.3)	137.3 (117.4–175.2)	126.9 (105.7–157.1)	0.076
IgM (g/L)	0.85 (0.78–1.01)	0.73 (0.63–0.91)	0.84 (0.75–0.95)	0.0067
IL-6 (pg/ml)	80 (68–91.08)	93.01 (81.01–100.9)	83.13 (72.8–94.26)	0.0114
IL-10 (pg/ml)	83.72 (70.76–97.9)	94.44 (80.86–104.4)	87.6 (75.33–100.6)	0.1093
CD100 ⁺ /CD4 ⁺ T cells %	66.4 (54.9–75.6)	53.9 (46.4–65.5)	61.8 (50.1–71.5)	0.0174
CD100 ⁺ /CD8 ⁺ T cells %	73.3 (63.1–77.9)	54.4 (48.9–69)	70.8 (56.2–76.3)	0.0003
CD72 ⁺ /CD19 ⁺ B cells %	68.4 (59.1–74.5)	54.9 (46–66.4)	65.8 (55.6–73.2)	0.0018

Data are shown as median (interquartile ranges), frequencies and percentages. The comparisons just performed between survivors and non-survivors, using Student's *t* test or Mann-Whitney *U* test. NA Not applicable, WBC Counts of white blood cell, LC Counts of lymphocyte, SOFA Sequential organ failure assessment, APACHE II Acute physiology and chronic health evaluation II, SAPS II Simplified acute physiology score II, CRP C-reaction protein, IgM Immunoglobulin M, IL interleukin



observed when compared septic non-survivors with survivors (Table 2). Furthermore, we found that lymphocyte counts were positively correlated with the percentages of CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, and CD72⁺/CD19⁺ B cells ($r = 0.5106, 0.2705, \text{ and } 0.3051$, respectively) (Fig. 3a, e, and i).

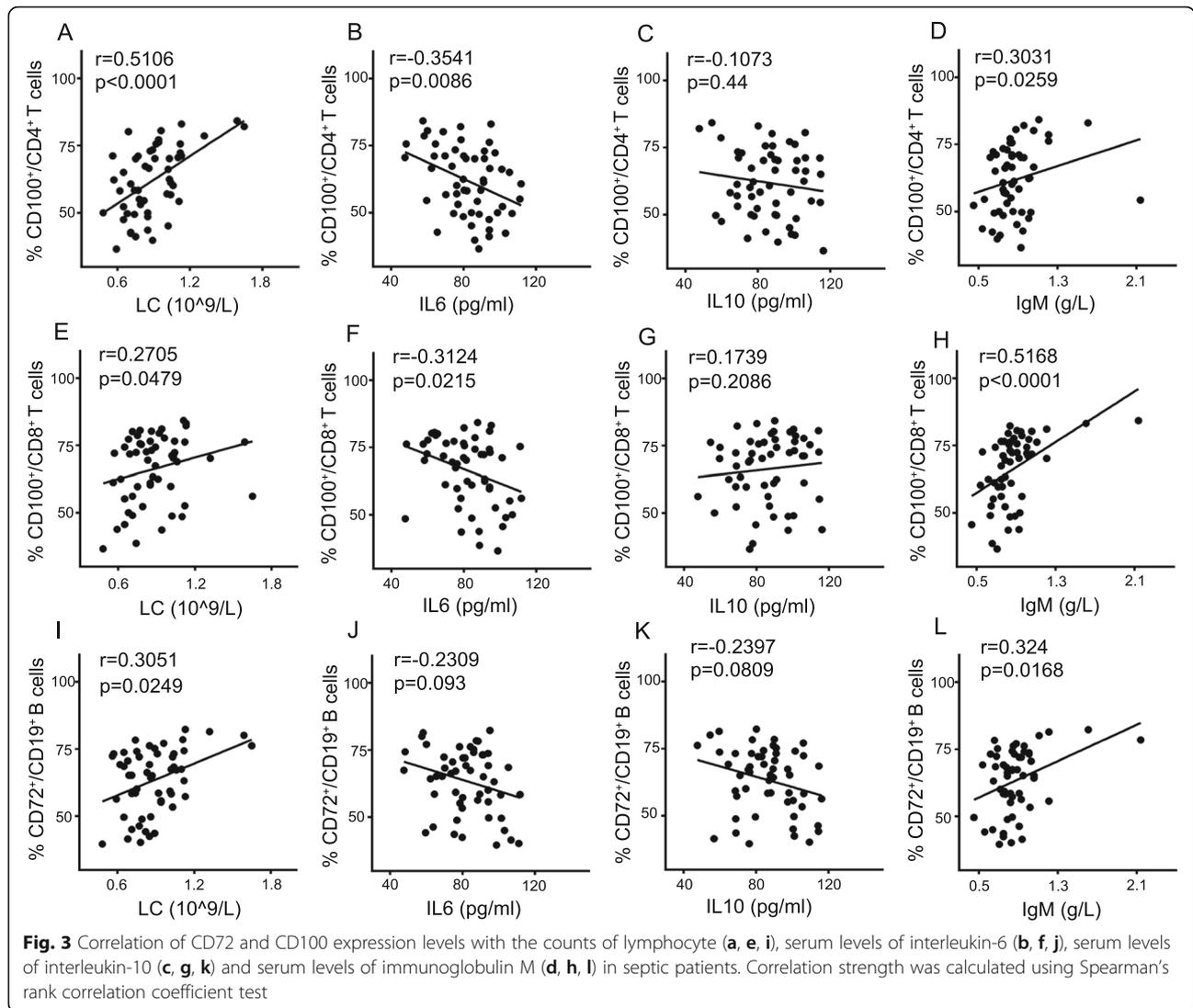
Predictive value of CD72/CD100-related molecules for mortality in septic patients

The ROC curves of CD72/CD100-related molecules for predicting 28-day mortality are shown in Fig. 4. The ROC curve analysis (area under the curve (AUC)) showed that the percentage of CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, CD72⁺/CD19⁺ B cells for predicting 28-day mortality were 0.695, 0.803, and 0.765, respectively (Table S3). It was clear that the levels of CD100 expression on CD8⁺ T cells had a better predictive value than the others.

Subsequently, we explored the prognostic significance of the combination of the best CD72- and CD100-related predictors (namely the levels of CD100 expression on CD8⁺ T cells) with conventional clinical risk parameters (SOFA score, APACHE II score, and SAPS II). The results displayed the combination of the percentage

of CD100⁺/CD8⁺ T cells and SOFA score had the largest AUC (Table S3). We estimated the difference of ROC between the single parameter and combined parameter by using DeLong test (Table S4). Our results showed that there were no statistical differences between the single parameter and combined parameter, though the combined parameter had larger AUCs than a single parameter.

Furthermore, by using cutoff values determined by ROC, we observed septic patients with the percentage of CD100⁺/CD4⁺ T cells, CD100⁺/CD8⁺ T cells, CD72⁺/CD19⁺ B cells lower than 54.9% (sensitivity was 76.3% and specificity was 56.3%), 62.9% (sensitivity was 78.9% and specificity was 75%), and 59.0% (sensitivity was 76.3% and specificity was 68.8%), respectively, had a lower probability of survival at day 28 after the onset of sepsis (Fig. 5). Univariate and multivariate logistic regression analysis were conducted to predict the 28-day mortality of patients with sepsis. At first, we conducted the univariate logistic regression analysis. Statistically significant variables in univariate analysis were conserved in the multivariate logistic model. Subsequently, the percentage of CD100⁺/CD8⁺ T cells ($B = -0.095, OR = 0.909, P = 0.004$) and the percentage of CD72⁺/CD19⁺



B cells ($B = -0.086$, $OR = 0.918$, $P = 0.012$) were found to be independent predictors of 28-day mortality in septic patients. The details were presented in Table 3.

Discussion

Sepsis is an important medical problem of the 21st century that warrant renewed attention. In China, the incidence of severe sepsis was estimated to be more than 100 cases per 100,000 people, and the hospital mortality of sepsis was nearly 20.6% [22]. The high mortality rates of sepsis are partially due to the lack of an effective approach to predict the prognosis of sepsis. Thus, identifying novel efficient biomarkers for a better management of sepsis is urgent. The current study firstly assessed the levels of CD72 and CD100 expression on circulating lymphocytes at days 3–4 after the onset of sepsis, resulting in three main findings. First, CD72 and CD100 expression levels were both significantly lower in septic

patients than in healthy controls. Second, the reduction of lymphocyte counts and serum IgM levels were related to the low expression levels of CD72 and CD100. Third, the percentage of CD100⁺/CD8⁺ T cells and CD72⁺/CD19⁺ B cells were both independent predictors of 28-day mortality in septic patients.

Lymphopenia is common in sepsis, both in non-survivors and survivors, and predicts poor prognosis [23, 24]. To date, numerous studies have revealed that CD100 could regulate the activation and proliferation of T cells via the interaction with CD72 [15]. Additionally, previous studies have proved that the expression of CD100 on circulating T cells was significantly decreased in HIV patients [25]. Similarly, authors also observed hepatitis C virus infection reduced CD100 expression on CD8⁺ T cells [26]. Moreover, accumulated evidence indicated that CD100 could rescue B cells from apoptosis and increase B-cell proliferation and survival [27, 28]. In

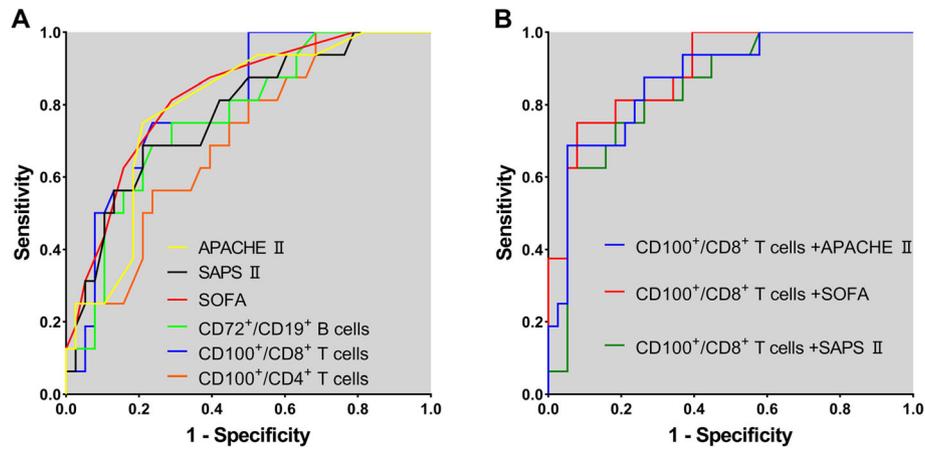


Fig. 4 Receiver operating characteristic (ROC) curves of various indicators in predicting 28-day mortality risk of septic patients. **a** The AUCs for individual parameter were 0.823 for SOFA score, 0.775 for SAPS II, 0.792 for APACHE II score, 0.695 for the percentage of CD100⁺/CD4⁺ T cells, 0.803 for the percentage of CD100⁺/CD8⁺ T cells, 0.765 for the percentage of CD72⁺/CD19⁺ B cells. **b** The AUCs for the combination of the percentage of CD100⁺/CD8⁺ T cells with SOFA score, SAPS II, or APACHE II score were 0.895, 0.846, and 0.872, respectively

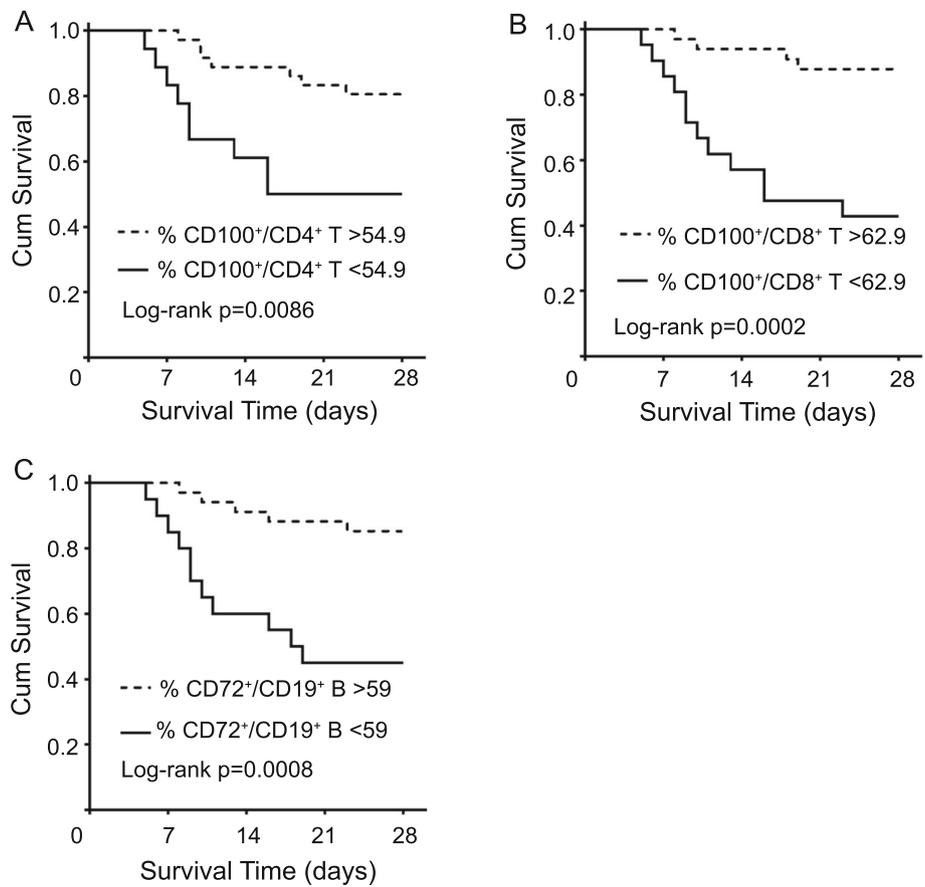


Fig. 5 Survival curves of septic patients according to the percentage of CD100⁺/CD4⁺ T cells (**a**), CD100⁺/CD8⁺ T cells (**b**), and CD72⁺/CD19⁺ B cells (**c**)

Table 3 Logistic regression analysis for prediction of 28-day mortality in sepsis

Variable	Univariate analysis			Multivariate analysis		
	B	OR (95%CI)	P	B	OR (95%CI)	P
CD100 ⁺ /CD4 ⁺ T cells	-0.06	0.941 (0.894–0.992)	0.023			
CD100 ⁺ /CD8 ⁺ T cells	-0.095	0.91 (0.859–0.963)	0.001	-0.095	0.909 (0.853v0.969)	0.004
CD72 ⁺ /CD19 ⁺ B cells	-0.084	0.92 (0.869–0.974)	0.004	-0.086	0.918 (0.858–0.982)	0.012

Statistically significant variables in univariate analysis were conserved in multivariate analysis (Forward, LR, $\alpha = 0.05$). B: logistic regression coefficient, OR: odds ratio. CI: confidence interval

line with the preceding research, our study showed the numbers of circulating lymphocyte were substantially decreased with decreasing CD72 and CD100 expression levels.

Lymphocyte exhaustion was recently recognized as a major mechanism of immune suppression in sepsis, and it was characterized by impaired lymphocyte activation, proliferation, IgM production, and increased susceptibility to secondary infection and poor survival in the immunosuppressive phase of sepsis [29, 30]. Moreover, low serum IgM levels also play a critical role in secondary infection during sepsis, and it is often correlated with increased mortality [30–32]. In this study, we also observed a significant reduction in serum IgM levels in septic patients. More importantly, we found the serum IgM levels were positively correlated with the expression levels of CD72 and CD100, especially the levels of CD100 expression on CD8⁺ T cells. This can be partly explained by the ligation of CD100 and CD72 could enhance the activation of B cells in antibody production [7]. Simultaneously, murine studies demonstrated the proliferative responses and immunoglobulin production of B cells were both significantly reduced in CD100-deficient mice [33].

Another major characteristic of lymphocyte exhaustion is the increase of expression of various inhibitory receptors, such as cytotoxic T-lymphocyte-associated protein 4 (CTLA-4), B and T lymphocyte attenuator (BTLA), Programmed cell death receptor-1 (PD-1), and Programmed cell death receptor ligand-1 (PD-L1) [34]. Furthermore, it is currently believed that PD-1 and PD-L1 both play important roles in sepsis-induced immunosuppression [21]. Recently, Correa et al. demonstrated that CD100 expression on T cells was associated with the expression of PD-1 and PD-L1 on T cells during HIV infection, meanwhile, CD72/CD100 axis was correlated with T-cell exhaustion [17]. Taken together, it is plausible to speculate that CD72 and CD100 may play major roles in the progression of sepsis by regulating lymphocyte exhaustion.

In theory, as an inhibitory receptor that expressed primarily on B cells, the expression levels of CD72 should be increased in varying degrees. However, in the current study, the CD72 expression showed a significant

decrease. It may be due to CD72 plays a major role at multiple stages of B-cell development and is necessary for establishing and maintaining the normal peripheral, conventional B-cell pool [14]. According to the development of B cells, B cells are typically classified into pro-B cells, pre-B cells, immature B cells, and mature B cells [35]. Simultaneously, CD72 expression is detectable throughout B-cell differentiation from early progenitors to mature B cells [14]. In adult CD72^{-/-} mice, the numbers of pro-B cells, pre-B cells, immature B cells, and mature B cells were normal, significant increase, minimally changed, and significant decrease, respectively [14, 36]. It indicated that the absence of CD72 diminishes the efficiency of the transitions from pre-B to immature B and from immature to mature B cells. Recently, Duan et al. also released that impaired B-cell maturation contributed to the reduction of B cell numbers, serum IgM levels and poor prognosis in sepsis [37]. Thus, we hypothesize the reduction of CD72 expression on B cells still plays a major role in the development of sepsis. However, more further studies are required in order to explore the alteration of CD72 expression in different stages of B cells.

Some limitations need to be considered in our study. First, the numbers of patients and healthy controls were relatively small, and the data were only collected at a single institution. Second, the expression of CD72/CD100 was only determined at a single time point, because it was difficult to obtain enough blood samples from all participants. Dynamic monitoring reflects the evolution of CD72/CD100 expression on lymphocytes during sepsis and it may yield predictors with better predictive value; thus, dynamic detection is essential for further study. Third, the number of female patients is too small to rule out the interference caused by gender. Furthermore, the age of septic patients was concentrated between 45 and 56 years; it remains to be explored whether such a conclusion could be generalized to other age groups.

Conclusions

To conclude, our study revealed that the decrease of the levels of CD72 and CD100 expression on circulating lymphocytes after 3–4 days of sepsis had a close correlation of the 28-day mortality of septic patients,

substantializing CD72 and CD100 as potential prognostic markers for sepsis.

Supplementary information

Supplementary information accompanies this paper at <https://doi.org/10.1186/s40560-020-00486-9>.

Additional file 1: Table S1. Baseline clinical characteristics of infection in septic patients.

Additional file 2: Table S2. Alterations of different lymphocyte subsets.

Additional file 3: Table S3. Area under the curve of various parameters for predicting 28-day mortality in septic patients.

Additional file 4: Table S4. The difference of ROC between the single parameter and combined parameter.

Abbreviations

WBC: The counts of white blood cell; LC: The counts of lymphocyte; CRP: C-reaction protein; SOFA: Sequential organ failure assessment; SAPS II: Simplified acute physiology score II; APACHE II: Acute physiology and chronic health evaluation II; SSC: Side scatter; FSC: Forward scatter; IL: Interleukin; IgM: Immunoglobulin M; ROC: Receiver operating characteristic; AUC: Area under the curve; BTLA: B and T lymphocyte attenuator; CTLA-4: Cytotoxic T-lymphocyte-associated protein 4; PD-1: Programmed cell death receptor-1; PD-L1: Programmed cell death receptor ligand-1

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Authors' contributions

FY is the guarantor of the article, taking responsibility for the integrity of the work as a whole, from inception to published article. XB and ZL supervised the project and provided research funding. JC collected and analyzed data. MT collected data and samples. CY critically revised the manuscript. WG helped data management. YY conceived the idea, performed flow cytometry experiment, drafted and revised the manuscript. All authors read and approved the final manuscript.

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Availability of data and materials

Data are available on request.

Ethics approval and consent to participate

This research was approved by the medical ethics committee of Tongji Hospital affiliated to Tongji Medical College of Huazhong University of Science and Technology, and informed consent was obtained from all participants according to the Declaration of Helsinki.

Consent for publication

Not applicable.

Competing interests

The authors declare that they have no conflicts of interest.

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