ORIGINAL ARTICLE

Serial Trend of Neutrophil CD64, C-reactive Protein, and Procalcitonin as a Prognostic Marker in Critically Ill Patients with Sepsis/Septic Shock: A Prospective Observational Study from a Tertiary Care ICU

Rupali Patnaik[1], Afzal Azim[2], Kritika Singh[3], Vikas Agarwal[4], Prabhaker Mishra[5], Banani Poddar[6], Mohan Gurjar[7], Shakti B Mishra[8]

ABSTRACT

Aim and background: Neutrophil CD64 (nCD64) is evolving as a prognostic biomarker in sepsis. The primary objective of this study was to evaluate whether serial trend of nCD64, procalcitonin (PCT), and C-reactive protein (CRP) predict 28-day mortality in patients with sepsis/septic shock, as per Sepsis-3 criteria.

Materials and methods: This prospective, observational single-center cohort study included 60 adult patients (age ≥18 years) with sepsis. Serial biomarker levels with SOFA score were measured at admission (day 0), on day 4, and on day 8.

Results: Of the 60 patients, 42 (70%) had septic shock. Biomarker levels at admission did not differ between patients with sepsis and septic shock. Thirty-seven patients survived and 23 were non-survivors by day 28. There was a significant fall in serial trend of all three biomarkers from admission till day 8 (Friedman $p < 0.001$) in survivors compared to a non-significant change in non-survivors. On multivariate analysis, SOFA score at admission (OR 1.731), more days with vasopressor support (OR 1.077), rise in CD64 from day 0 to day 8 (OR 1.074), and rise in CRP from day 0 to 8 (OR 1.245) were the significant predictors of 28-day mortality ($p < 0.05$). The highest area under the ROC curve was obtained for more days of vasopressor therapy (0.857), followed by a rise in CD64 from day 0 to day 8 (0.798).

Conclusion: Serial trend of biomarkers has prognostic utility. The rise in CD64 from day 0 to day 8 was a good predictor of mortality compared to the trend of other biomarkers.

Keywords: C-reactive protein, Mortality, Neutrophil CD64, Procalcitonin, Sepsis, Septic shock, Survivor.

Indian Journal of Critical Care Medicine (2024): 10.5005/jp-journals-10071-24777

HIGHLIGHTS

A rise in nCD64 from admission to day 8 is a strong predictor of mortality, underlining the potential clinical value of monitoring this biomarker in sepsis/septic shock patients. Our study highlights that if there is an increase in nCD64 still by day 8 compared to baseline, then clinicians should review for appropriate therapeutic intervention or modifications.

INTRODUCTION

Sepsis and septic shock are substantial medical concerns that result in multiple organ dysfunction and high fatality rates among critically ill patients getting admission to intensive care units (ICUs). Although medical care has made significant progress, prompt identification, and suitable treatment interventions continue to be difficult due to the aggressive nature of these conditions.[1,2] One of the key challenges is the precise identification of individuals who have a significant likelihood of death and could potentially benefit from additional monitoring and therapeutic interventions.[2]

Biomarkers have become an appealing tool for the management of sepsis, with potential applications in the areas of diagnosis, prognosis, and therapeutic guiding.[3–6] Extensive research has been conducted on C-reactive protein (CRP) and procalcitonin

[1,8]Department of Critical Care Medicine, IMS and SUM Hospital, Bhubaneswar, Odisha, India

[2,6,7]Department of Critical Care Medicine, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India

[3,4]Department of Clinical Immunology and Rheumatology, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India

[5]Department of Biostatistics, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India

Corresponding Author: Afzal Azim, Department of Critical Care Medicine, Sanjay Gandhi Postgraduate Institute of Medical Sciences, Lucknow, Uttar Pradesh, India, Phone: +91 9984988393, e-mail: draazim2002@gmail.com

(PCT) in sepsis, focusing on their use in diagnosing and predicting outcomes.[7–11] However, there is a rising interest in exploring the potential of neutrophil CD64 (nCD64) as a biomarker for prognosis. Neutrophil CD64 is an immunoglobulin FcγR1 receptor with a strong binding affinity. It is expressed at higher levels in neutrophils as a response to infection.[12–14] Neutrophil CD64 is a well-established biomarker used for diagnosing sepsis, as supported by multiple studies.[3–7,15–19]

While prior research has explored the predictive significance of nCD64 in sepsis,[20–24] only a few number of studies have assessed the consequences of its sequential pattern.[25–27] Hence, the purpose of this study is to systematically identify the predictive significance of the sequential pattern of nCD64, in addition to PCT and CRP, in individuals diagnosed with sepsis and septic shock according to Sepsis-3 criteria.

We hypothesize that a substantial alteration in the sequential pattern of nCD64 levels can serve as a predictive indicator for 28-day mortality in patients diagnosed with sepsis and septic shock. This research has the potential to enhance our comprehension of the significance of nCD64 in predicting the outcome of sepsis and maybe provide guidance for future therapeutic approaches. This prospective observational cohort study was performed to assess the predictive value of changes in biomarkers over time in patients diagnosed with sepsis according to the Sepsis-3 criteria. The main aim of this study was to assess whether the sequential pattern of nCD64, PCT, and CRP biomarkers can predict the death rate within 28 days in our group of participants.

MATERIALS AND METHODS

Research Methodology and Location

The study was a prospective, observational, single-center cohort study undertaken in a 20-bed, adult mixed medical-surgical ICU in a 1,200-bed tertiary care institute in Northern India from January 2018 to December 2019.

Sample Size Calculation and Patient Selection

The entire sample size was determined using the receiver operating characteristic (ROC) curve formula. Prior research indicated that the ratio of survival to death among sepsis patients in ICUs was 2:1, and the estimated area under the ROC curves (AUC) of nCD64 in predicting mortality was 0.727. Therefore, employing MedCalc version 20.305 with a power of 90%, an alpha error of 0.05, and a default null hypothesis value of 0.5, the minimum required sample size was determined to be 72. This included a minimum of 48 survivors and a minimum of 24 non-survivors.

All sepsis patients with an identified infection source and who met the diagnostic criteria for sepsis based on the third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3) were invited to participate in the study. The study excluded patients who were under the age of 18, had cancer, declined to provide informed consent, or passed away within 48 hours after being admitted to the ICU. The study received approval from the institutional ethics committee, namely the IEC No: 2018-20-DM-102, at the Sanjay Gandhi Postgraduate Institute of Medical Sciences. Additionally, registration was done with the clinical trial registry of India under the code CTRI/2020/04/024795. Informed written consent was obtained from either the patients or their legal representatives in cases where the patients' health state rendered them unable to provide informed consent.

Data Collection and Biomarker Measurement

At ICU admission, a survey was administered to gather initial data from the participants. This data included demographic details such as age, sex, and BMI, as well as information on any existing health conditions. Additionally, clinical indicators such as blood pressure, respiratory rate, and pulse rate were recorded. The severity of illness was assessed using the SOFA and APACHE II scores, and the participants were categorized into different stages of sepsis (sepsis and septic shock). The laboratory tests conducted included a comprehensive blood count which involved measuring the white blood cell (WBC) count, coagulation test, serum lactate, PCT, CRP, and the nCD64 marker.

Upon admission, clinical parameters and blood samples were obtained. Subsequently, additional collections were made on day 4 and day 8. The laboratory tests were conducted at the standardized laboratory department of our institution. Flow cytometry was used to analyze nCD64, and the levels were represented as the percentage of neutrophils that showed CD64 positivity. Serum CRP and PCT levels were measured using nephelometry and electrochemiluminescence immunoassay, respectively.

Neutrophil CD64 Staining

Neutrophil CD-64 was analyzed as per protocol already established in our lab (Clin Rheumatol. 2019 Apr;38(4):997–1005) with slight modifications. Briefly, to 100 µL of fresh EDTA blood, 5µL of phycoerythrin (PE) labeled mouse anti-Human CD-64 antibody (Cat no: 558592, BD Pharmingen™) was introduced and allowed to incubate at ambient temperature for 30 minutes in dark. A similar process was repeated in another tube with 100 µL of fresh EDTA blood and allowed to incubate with PE-conjugated matched-isotype control antibody. Red blood corpuscles' lysis was carried out with 1mL of 1X BD FACS™ Lysing solution at ambient temperature.

The tubes were washed for two rounds with 1X PBS at a centrifugal speed of 1200 rpm for 8 minutes. The liquid portion was discarded and the solid portion was mixed again in a solution of 1X PBS. The acquisition was performed using the BD FACSCanto™II flow cytometer (Becton Dickinson, San Jose, California, USA). A total of 10,000 cells were enumerated and examined utilizing BDFACS DIVA software version 8.0. The neutrophil population was selected based on the forward and side scatter characteristics, as shown in Figure 1. The identification of a cell population with positive staining for the CD64 marker was achieved by comparing dual-parameter histograms to the equivalent isotype control, with the mean fluorescence intensity (MFI) serving as an indicator. The results were quantified by calculating the percentage of neutrophils expressing CD64 and by measuring the MFI.[28]

Statistical Assessment

The presentation of continuous data included the use of median and interquartile range (IQR), while categorical variables were expressed in terms of frequency and percentage. The Mann-Whitney U test was used to examine the distributions of continuous variables between patients who survived and those who did not, whereas the Chi-square test was used for categorical variables. The Friedman test was employed to examine changes in distributions over time, and subsequent multiple comparisons were conducted. Receiver operating characteristic curves were employed to ascertain the threshold values of the predictors for mortality. The statistical analyses were conducted using SPSS version 23 and R Statistical Software (version 3.6.2). A *p*-value below 0.05 was deemed
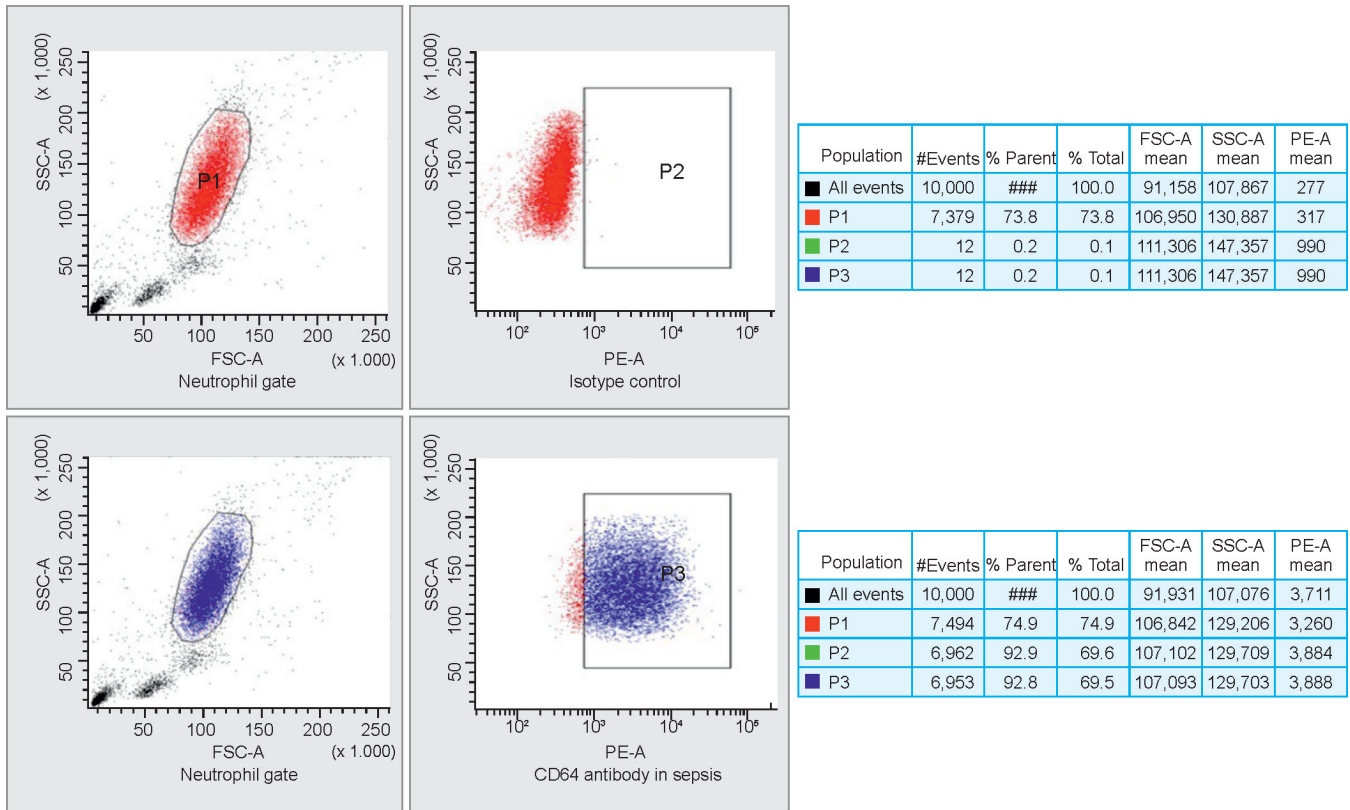
Population	#Events	% Parent	% Total	FSC-A mean	SSC-A mean	PE-A mean
■ All events	10,000	###	100.0	91,158	107,867	277
■ P1	7,379	73.8	73.8	106,950	130,887	317
■ P2	12	0.2	0.1	111,306	147,357	990
■ P3	12	0.2	0.1	111,306	147,357	990

Population	#Events	% Parent	% Total	FSC-A mean	SSC-A mean	PE-A mean
■ All events	10,000	###	100.0	91,931	107,076	3,711
■ P1	7,494	74.9	74.9	106,842	129,206	3,260
■ P2	6,962	92.9	69.6	107,102	129,709	3,884
■ P3	6,953	92.8	69.5	107,093	129,703	3,888

Fig. 1: Neutrophil gating strategy

Neutrophils were gated (P1) as per forward and side scatter characteristics. Tube with matched isotype antibody showing only 0.1% cells (P2) to be positive (right upper panel) whereas tube with neutrophil CD64 antibody showing 92.9% (P3) cells to be positive (right lower panel)

statistically significant. In order to assess the ability of changes in nCD64 values over time to predict mortality in ICU patients, we conducted a multivariate logistic regression analysis. This study considered potential confounding factors such as age, sex, and SOFA score. The odds ratios and 95% confidence intervals were computed to evaluate the magnitude and direction of the associations.

In addition, a Cox proportional hazards model was employed to assess the combined impact of numerous determinants on survival time. The hazard ratios and their corresponding 95% confidence intervals were computed. The validity of the proportional hazards assumption was assessed by examining Schoenfeld residuals.

Finally, in order to evaluate the models' ability to forecast accurately, the area under the ROC curve (AUC) was computed. The ROC curve with an AUC value of 0.5 signifies that the model has no discriminatory power, meaning it is no more accurate than random chance in predicting the outcome. Conversely, an AUC value of 1.0 signifies flawless discrimination, indicating that the model can perfectly predict the event. The DeLong test was employed to assess and compare the AUC values of various models.

The statistical tests conducted were two-sided, and a p-value below 0.05 was deemed to be significant statistically.

RESULTS

During the study period (January 2018 to December 2019), 123 patients were admitted to the ICU with sepsis and 78 patients fulfilled the inclusion criteria. Data of 18 participants could not be analyzed as they died within 8 days of admission to ICU. Flowchart of

study enrollment has been shown in Figure 2. The duration between ICU admission and the event of interest (death within 28 days) was considered as survival time. Those who did not experience the event during the study period were censored at the end of 28 days.

The demographic information, initial features, and admission biomarker levels of all 60 patients, including those with sepsis and septic shock are shown in Table 1. The median age of patients was 60 years, with 70% of them being male. The APACHE II score upon admission had a median value of 18, while the SOFA score had a median value of 8. The median period of illness prior to ICU admission was 4 days. The median length of stay in ICU was 9 days for individuals who survived and 7 days for those who did not survive. The mortality rate within 28 days was 28.3%.

Serial Trend of Biomarker Levels in 28-Day Survivors and Non-survivors

The serial trend of biomarker levels from day 0 to day 8 is presented in Figure 3. The median nCD64 level at admission was 91.0% in those who survived and 74.1% in those who did not survive. The median nCD64 level increased to 95.6% by day 4 in non survivors. By day 8, the median nCD64 level decreased to 69.2% in survivors whereas increased to 97.8% in non survivors. The disparity in nCD64 levels between survivors and non-survivors was statistically significant on day 8 ($p < 0.05$) (Table S1, Supplementary material).

Figure 4 displays the ROC curves that assess the ability of nCD64, CRP, and PCT levels on day 0, day 4, and day 8 to predict 28-day mortality. The AUC for nCD64 on day 8 was 0.727, surpassing the AUCs for both CRP and PCT. We attempted to determine the threshold value of CD64 for accurately predicting death. An

Table 1: Biomarker profile and clinical severity scores of patients admitted with sepsis/septic shock

Characteristics	All patients (n = 60)	Sepsis (n = 18)	Septic shock (n = 42)
Age, median (IQR)	35 (24–56)	31 (20–55)	43 (25–56)
Gender male, n (%)	37 (61.7%)	12 (66.7%)	25 (59.5%)
IBW, median (IQR)	68 (55–63)	62 (54–70)	63 (55–68)
No. of comorbidity, median (IQR)	2 (0–3)	1 (0–1)	1 (0–1)
APACHE II, median (IQR)	18 (12–22)	13 (12–17)	18 (16–22)
SOFA, median (IQR)	9 (6–12)	6 (4–10)	10 (7–12)
Admission category, n (%)			
Medical	53 (88.3%)	15 (83.3%)	38 (90.5%)
Surgical	7 (11.7%)	3 (16.7%)	4 (9.5%)
Days of illness before ICU admission, median (IQR)	9 (6–12)	7 (4–10)	10 (3–17)
Location prior to ICU admission, n (%)			
In hospital emergency	24 (40%)	8 (44.44%)	16 (38.09%)
In hospital ward	19 (31.66%)	6 (33.33%)	13 (30.95%)
In hospital ICU	4 (6.66%)	1 (5.55%)	3 (7.14%)
Outside ICU	13 (21.66%)	5 (27.7%)	8 (19.04%)
Culture positive at admission, n (%)	27 (45%)	8 (44.44%)	19 (45.23%)
Community/nosocomial source of sepsis	20/40	8/10	12/30
Comorbidities, n (%)			
Diabetes	11 (18.33%)	3 (16.66%)	8 (19.04%)
Hypertension	12 (20%)	3 (16.66%)	9 (21.42%)
Hypothyroid	10 (16.66%)	4 (22.22%)	6 (14.28%)
CAD	6 (10%)	2 (11.11%)	4 (9.52%)
Chronic resp. illness	5 (8.33%)	1 (5.55%)	4 (9.52%)
Liver disease	7 (11.66%)	2 (11.11%)	5 (11.90%)
ESRD	7 (11.66%)	1 (5.55%)	6 (14.28%)
Rheumatologic disorder	9 (15%)	2 (11.11%)	7 (16.66%)
Obesity	5 (8.33%)	2 (11.11%)	3 (7.14%)
None	41 (68.33%)	12 (66.66%)	29 (69.04%)
Diagnostic category, n (%)			
Pneumonia	16 (26.66%)	3 (16.66%)	13 (30.95%)
Severe acute pancreatitis	11 (18.33%)	4 (22.22%)	7 (16.66%)
Tropical infection	8 (13.33%)	2 (11.11%)	6 (14.28%)
Gullaine-Barre syndrome	7 (11.66%)	3 (16.66%)	4 (2.38%)
Stroke and meningoencephalitis	5 (8.33%)	2 (11.11%)	3 (7.14%)
GI condition	5 (8.33%)	2 (11.11%)	3 (7.14%)
ALF/ACLF	4 (6.66%)	1 (5.55%)	3 (7.14%)
Urinary tract infection	2 (3.33%)	1 (5.55%)	1 (2.38%)
Skin and subcutaneous tissue infection	2 (3.33%)	1 (5.55%)	1 (2.38%)
Organ system involved, n (%)			
Respiratory	52 (86.66%)	15 (83.33%)	37 (88.09%)
Cardiovascular	42 (70.00%)	13 (72.22%)	29 (69.04%)
Renal	35 (58.33%)	9 (50%)	26 (61.90%)
Abdominal	17 (28.33%)	5 (27.7%)	12 (28.57%)
Neurological	16 (26.66%)	4 (22.22%)	12 (28.57%)
Hematological	8 (13.33%)	2 (11.11%)	6 (14.28%)
Biomarker levels, median (IQR)			
Neutrophil CD64 (%)	90.8 (70.0–98.68)	81.7 (53.2–94.75)	91.65 (70–99.02)
CRP (mg/dL)	11.7 (4.68–17.82)	8.5 (4.46–17.07)	12.65 (5.08–19.40)
PCT (ng/mL)	1.67 (0.89–4.39)	1.65 (0.39–2.98)	1.76 (1.17–19.40)

APACHE, acute physiology and chronic health evaluation; ACLF, acute on chronic liver failure; ALF, acute liver failure; CAD, coronary artery disease; CRP, C-reactive protein; ESRD, end stage renal disease; GI, gastro intestinal; IBW, ideal body weight; ICU, intensive care unit; IQR, interquartile range; SOFA, sequential organ failure assessment; PCT, procalcitonin; nCD64, neutrophil cluster of differentiation 64

increase of 11% in nCD64 from day 0 to day 8 was discovered to be a reliable indicator of death, with a sensitivity of 87.5% and a specificity of 58%.

In the multivariate logistic regression analysis, an increase in nCD64 level from day 0 to day 8 was a strong predictor of mortality at 28 days (odds ratio = 1.10, 95% confidence interval = 1.02–1.19, $p < 0.05$). The duration of vasopressor support was also a strong predictor of 28-day mortality (odds ratio = 1.09, 95% confidence interval = 1.01–1.18, $p < 0.05$) (Table 2).

Serial Biomarker Levels of Patients with Sepsis and Septic Shock at Admission

Out of a total of 60 patients, 42 (60%) had septic shock at admission. Median biomarker levels along with SOFA score at admission and on serial days were compared between patients admitted with sepsis and septic shock. Those admitted with septic shock had significantly

Fig. 2: Flowchart of study enrollment

notably elevated SOFA scores than those with sepsis at admission (day 0) and on day 4 ($p < 0.05$). The levels of all three biomarkers showed no significant differences between patients admitted with septic shock and those with sepsis on day 0, day 4, and day 8. See Figure S1, supplementary material.

DISCUSSION

Our study provides novel insights into the prognostic utility of serial measurements of nCD64 expression in individuals with sepsis and septic shock, according to the Sepsis-3 criteria. Our research revealed that a rise in nCD64 levels from the initial measurement to day 8 was a strong indicator of the likelihood of death within 28 days. This suggests that measuring nCD64 levels could be useful in assessing the risk and treatment of these patients. The role of nCD64 as a biomarker in sepsis and septic shock has gained considerable interest in recent years. Further, CD64, or FcγRI, is a receptor with a strong attraction for the Fc portion of IgG and is found on the outer layer of neutrophils. Also, CD64 expression on neutrophils is increased during infection and inflammation, making it a potential marker of immune activation in sepsis and septic shock.[14,15]

The findings of our study are in accordance with previous literature demonstrating the prognostic significance of nCD64 in sepsis and septic shock.[20–27] For instance, studies have found that nCD64 was notably higher in sepsis patients who did not survive compared to those who did, and this expression was associated
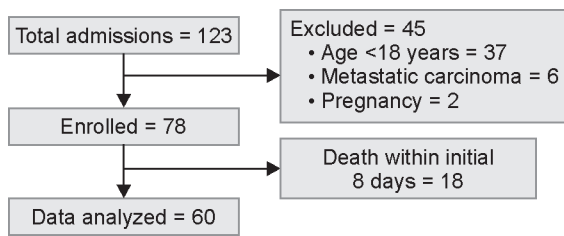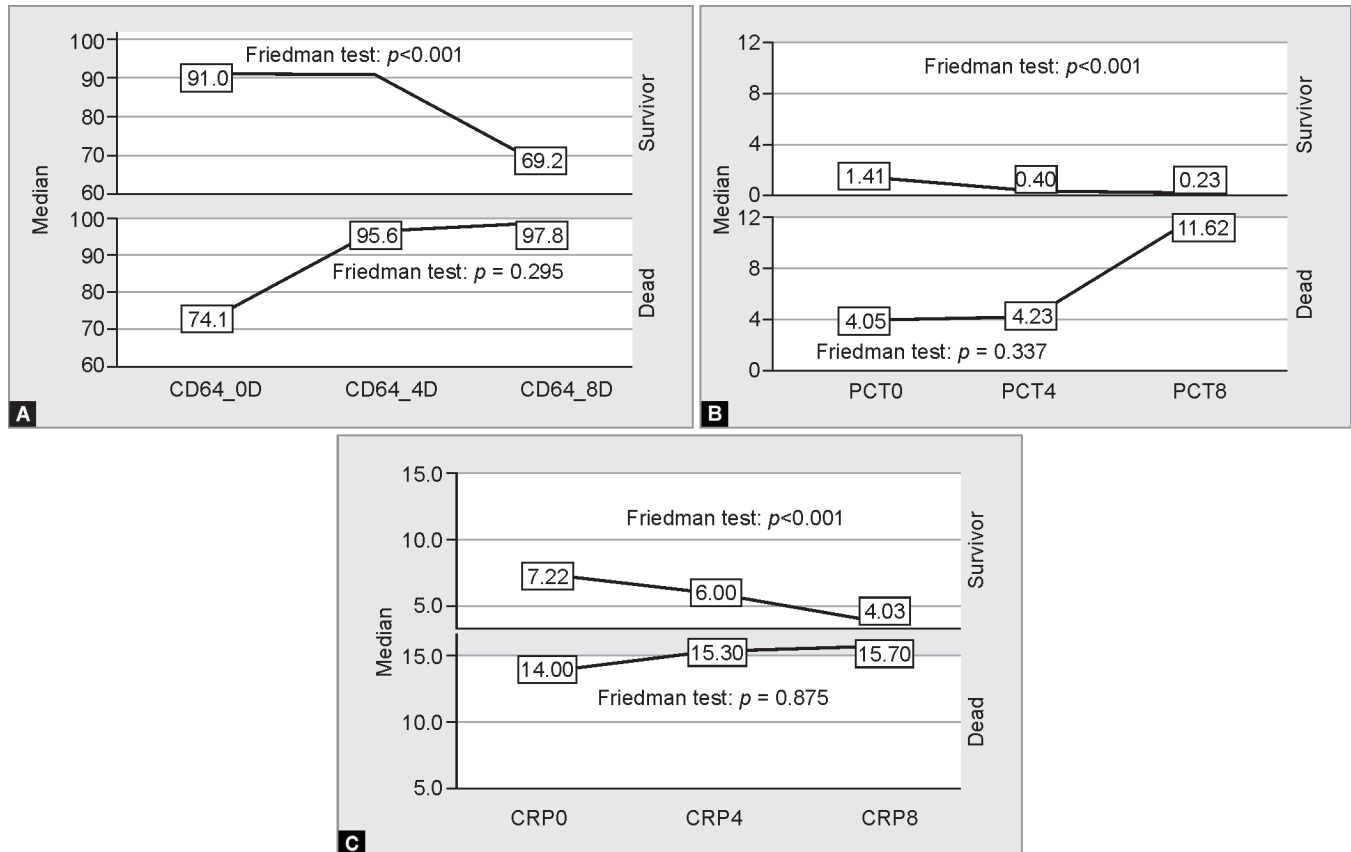
Figs 3A to C: Comparison between 28-day survivors and non-survivors of serial trend of neutrophil CD64, PCT, and CRP, respectively. nCD64, cluster of differentiation factor 64, CRP, C-reactive protein PCT, procalcitonin

with the severity of the disease.[20,24,25,27] Similarly, studies have reported that nCD64 expression was a significant predictor of death in patients with septic shock.[21–24,26]

However, our study is unique in that it prospectively evaluated the serial trend of nCD64, along with CRP and PCT, in a cohort of patients defined by Sepsis-3 criteria. This approach allowed us to capture dynamic changes in these biomarkers over time and assess their predictive value at different stages of the disease.

Interestingly, we found that the prognostic significance of nCD64 was superior to that of CRP and PCT, which are widely used biomarkers in sepsis and septic shock. This finding is in line with the results of a recent study by Huang et al., which reported that nCD64 had a higher area under the ROC curve (AUC) for predicting mortality in sepsis patients compared to CRP and PCT.[23]
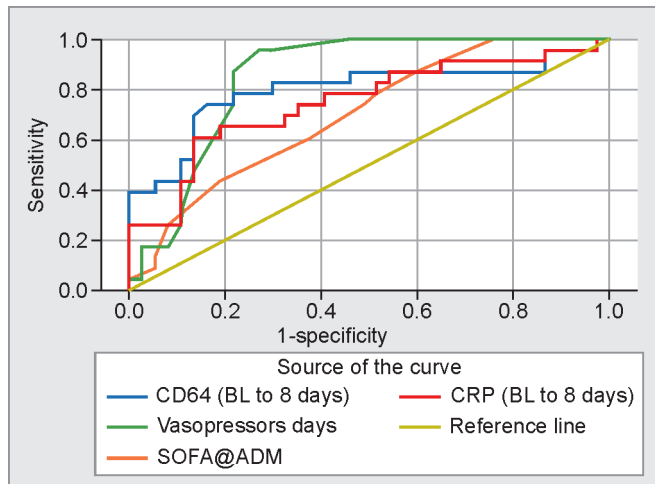

Fig. 4: ROC curve analysis of predictors of 28-day mortality on multivariate analysis

CD64 (BL to 8 days) = rise in nCD64 from baseline (day 0) to 8 days, CRP (BL to 8 days) = rise in CRP from baseline (day 0) to 8 days, SOFA@ADM, admission SOFA score, CRP, C-reactive protein, PCT, procalcitonin, SOFA, sequential organ function assessment, nCD64, cluster of differentiation factor 64

The superior performance of nCD64 could be attributed to its contribution to the immune response to infection. Unlike CRP and PCT, which are acute-phase proteins produced in response to inflammation, nCD64 is directly involved in the immune response to infection. It mediates the phagocytosis of pathogens and the release of pro-inflammatory cytokines, making it a more specific marker of immune activation in sepsis and septic shock. Our study also revealed that patients requiring longer durations of vasopressor support had higher mortality rates, underscoring the importance of hemodynamic stability in the prognosis of sepsis and septic shock. This finding is in accordance with previous literature that has identified vasopressor requirement as a key predictor of poor outcomes.[29]

Our study was conducted in a single center, which may limit the generalizability of the results. However, the center is a tertiary care hospital that treats a diverse patient population, increasing the likelihood that our findings are applicable to other settings. Furthermore, single-center studies are often the first step in investigating new research questions, and our study provides a valuable foundation for future multi-center studies. Since the sample size of our study was relatively small, it could limit the statistical power and the precision of our estimates. However, our sample size was determined based on a power calculation, and we were able to detect statistically significant differences in nCD64 levels between survivors and non-survivors. Though the sample size is small, our study provides preliminary evidence of the prognostic value of nCD64 in sepsis and septic shock. We excluded patients who died within 8 days of admission, which could potentially introduce bias. However, this decision was made to ensure that we had complete data for all included patients. Our findings are still relevant for the majority of patients with sepsis and septic shock, who survive beyond the initial phase of the illness. We did not validate the cut-off values for the biomarkers in an independent cohort, which could limit the reliability of these cut-off values in other patient populations or settings. However, our study is one of the first to investigate the serial trend of nCD64 in sepsis and septic shock, and our findings provide a starting point for determining optimal cut-off values. Future studies could validate these cut-off values in different cohorts. Although we controlled

Table 2: Univariate and multivariate analysis of predictors of 28-day mortality

	OR	95% CI	p-value	AOR	95% CI	p-value
Parameters	*Univariate analysis*			*Multivariate analysis*		
Admission APACHE II	1.184	1.054–1.133	0.005*			
Admission SOFA	1.262	1.056–1.507	0.010*	1.731	1.223–2.452	0.002*
Septic shock at admission	2.891	0.815–10.26	0.100			
↑SOFA (D0–D4)	1.129	0.987–1.506	0.066			
↑SOFA (D0–D8)	1.480	1.183–1.851	0.001*			
↑nCD64 (D0–D4)	1.009	0.987–1.013	0.448			
↑nCD64 (D0–D8)	1.049	1.017–1.081	0.002*	1.074	1.026–1.125	0.002*
↑PCT (D0–D4)	0.988	0.957–1.020	0.453			
↑PCT (D0–D8)	1.015	0.986–1.045	0.316			
↑CRP (D0–D4)	1.017	0.998–1.150	0.055			
↑CRP (D0–D8)	1.151	1.046–1.266	0.004*	1.245	1.048–1.476	0.013*
Days on vasopressor therapy	1.100	1.013–1.194	0.023*	1.077	1.002–1.157	0.043*

*$p < 0.05$ considered statistically significant. AOR, adjusted odds ratio; APACHE, acute physiology and chronic health evaluation; CI, confidence interval; CRP, C-reactive protein; OR, odds ratio; PCT, procalcitonin; SOFA, sequential organ failure assessment; nCD64, neutrophil cluster of differentiation 64; ↑, increase/rise

for some potential confounding factors in our analysis, there may be other unmeasured factors that could influence the relationship between the biomarkers and mortality. However, we included key variables known to affect the prognosis of sepsis and septic shock in our analysis, and our findings remained significant after adjusting for these variables. Our study only followed patients for 28 days, so it's unclear whether our findings apply to long-term outcomes. However, the 28-day mortality rate is a commonly used outcome measure in studies of sepsis and septic shock, and our findings provide important insights into the early prognosis of these patients.

Despite these limitations, this study makes a robust contribution to the literature by providing new insights into the prognostic value of nCD64 in sepsis and septic shock. Moreover, we assessed nCD64 levels serially till day 8, compared to others that measured this biomarker for 48 or 72 hours thereby its utility is solely for prognosis.[25,26] Our study highlights that if there is an increase in nCD64 still by day 8 compared to baseline, clinicians should consider appropriate therapeutic intervention or modifications. We believe that our findings warrant publication and will stimulate further research in this area.

Conclusion

Among critically ill patients with sepsis and septic shock, an increase in nCD64 levels from baseline to day 8 is a significant predictor of mortality. There is a need for further prospective studies with larger sample sizes to evaluate the prognostic value of serial nCD64 trends and explore the potential benefits of combining this biomarker with others to improve prognostic accuracy in this patient population.

Clinical Significance

A rise in nCD64 from admission to day 8 is a strong predictor of mortality, underlining the potential clinical value of monitoring this biomarker in sepsis/septic shock patients. If there is an increase in nCD64 still by day 8 compared to baseline, then clinicians should review for appropriate therapeutic intervention or modifications.

Declaration

Ethics Approval

The study was approved by the institutional ethics committee (IEC No: 2018-20-DM-102) of Sanjay Gandhi Postgraduate Institute of Medical Sciences and registered with the Clinical Trial Registry of India (CTRI/2020/04/024795).

Author Contributions

RP—Data curation; Formal analysis; Investigation; Methodology; Writing original draft; Writing—review and editing. AA—Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Project administration; Supervision; Writing original draft; Writing —review and editing. VA—Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Supervision; Writing—review and editing. KS—Data curation; Formal analysis; Investigation; Methodology; Sample processing and laboratory work administration. PM—Data curation; Formal analysis; Statistical analysis; Investigation; Methodology; Writing—review and editing. MG, BP, and SBM—Investigation; Methodology; Writing—review and editing.

Supplementary Materials

All the supplementary materials are available online on the website of www.IJCCM.org.

Orcid

Rupali Patnaik https://orcid.org/0000-0003-0104-7102
Afzal Azim https://orcid.org/0000-0003-3077-5424
Kritika Singh https://orcid.org/0000-0002-3814-4179
Vikas Agarwal https://orcid.org/0000-0002-4508-1233
Prabhaker Mishra https://orcid.org/0000-0003-4769-9106
Banani Poddar https://orcid.org/0000-0002-1843-3037
Mohan Gurjar https://orcid.org/0000-0002-8489-0324
Shakti B Mishra https://orcid.org/0000-0001-6634-1877

References

1. Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, et al. The third international consensus definitions for sepsis and septic shock (Sepsis-3) JAMA 2016;315(8):801–810. DOI: 10.1001/jama.2016.0287.
2. Rhodes A, Evans LE, Alhazzani W, Levy MM, Antonelli M, Ferrer R, et al. Surviving sepsis campaign: International guidelines for management of sepsis and septic shock 2016. Crit Care Med 2017;45:486–552. DOI: 10.1007/s00134-017-4683-6.
3. Pierrakos C, Vincent JL. Sepsis biomarkers: A review. Crit Care 2010;14(1):R15. DOI: 10.1186/cc8872.
4. Larsen FF, Petersen JA. Novel biomarkers for sepsis: A narrative review. Eur J Intern Med 2017;45:46–50. DOI: 10.1016/j.ejim.2017.09.030.
5. Pierrakos C, Velissaris D, Bisdorff M, Marshall JC, Vincent JL. Biomarkers of sepsis: Time for a reappraisal. Crit Care 2020;24(1):287. DOI: 10.1186/s13054-020-02993-5.
6. Liu Y, Hou JH, Li Q, Chen KJ, Wang SN, Wang JM. Biomarkers for diagnosis of sepsis in patients with systemic inflammatory response syndrome: A systematic review and meta-analysis. Springerplus 2016;5:2091. DOI: 10.1186/s40064-016-3591-5.
7. Yeh CF, Wu CC, Liu SH, Chen KF. Comparison of the accuracy of neutrophil CD64, procalcitonin, and C-reactive protein for sepsis identification: A systemic review and meta-analysis. Ann Intensive Care 2019;9(1):5. DOI: 10.1186/s13613-018-0479-2.
8. Garnacho-Montero J, Huici-Moreno MJ, Gutierrez-Pizarraya A, López I, Márquez-Vácaro JA, Macher H, et al. Prognostic and diagnostic value of eosinopenia, C-reactive protein, procalcitonin, and circulating cell-free DNA in critically ill patients admitted with suspicion of sepsis. Crit Care 2014;18:R116. DOI: 10.1186/cc13908.
9. Patnaik R, Azim A, Mishra P. Should serial monitoring of procalcitonin be done routinely in critically ill patients of ICU: A systematic review and meta-analysis. J Anaesthesiol Clin Pharmacol 2020;36:458–464. DOI: 10.4103/joacp.

10. Poddar B, Gurjar M, Singh S, Aggarwal A, Singh R, Azim A, Baronia A, et al. Procalcitonin kinetics as a prognostic marker in severe sepsis/septic shock. Indian J Crit Care Med 2015;19(3):140–146. DOI: 10.4103/0972-5229.152755.

11. Jamsa J, Ala-Kokko T, Huotari V, et al. Neutrophil CD64, C-reactive protein, and procalcitonin in the identification of sepsis in the ICU - Post-test probabilities. J Crit Care 2018;43:139–142, DOI: 10.1016/j.jcrc.2017.08.038.

12. Schiff DE, Rae J, Martin TR, Davis BH, Curnutte JT. Increased phagocyte Fc gamma RI expression and improved Fc gamma-receptor-mediated phagocytosis after in vivo recombinant human interferon-gamma treatment of normal human subjects. Blood 1997;90(8):3187–3194. DOI: 10.1182/blood.V90.8.3187.

13. Mangalam AK, Yadav R. Utility of CD64 expression on neutrophils as a marker to differentiate infectious versus non-infectious disease flares in autoimmune disorders. Indian J Rheumatol 2019;14(1):9–11. DOI: 10.4103/0973-3698.254192.

14. Hoffmann JJ. Neutrophil CD64: A diagnostic marker for infection and sepsis. Clin Chem Lab Med 2009;47(8):903–916. DOI: 10.1515/CCLM.2009.224.

15. Patnaik R, Azim A, Agarwal V. Neutrophil CD64 a diagnostic and prognostic marker of sepsis in adult critically ill patients: A brief review. Indian J Crit Care Med 2020;24(12):1242–1250. DOI: 10.5005/jp-journals-10071-23558.

16. Cid J, Aguinaco R, Sánchez R, García-Pardo G, Llorente A. Neutrophil CD64 expression as marker of bacterial infection: A systematic review and meta-analysis. J Infect 2010;60(5):313–319. DOI:10.1016/j.jinf.2010.02.013.

17. Li S, Huang X, Chen Z, Zhong H, Peng Q, Deng Y, et al. Neutrophil CD64 expression as a biomarker in the early diagnosis of bacterial infection: A meta-analysis. Int J Infect Dis 2013;17(1):e12–23. DOI: 10.1016/j.ijid.2012.07.017.

18. Wang X, Li ZY, Zeng L, Zhang AQ, Pan W, Gu W, et al. Neutrophil CD64 expression as a diagnostic marker for sepsis in adult patients: A meta-analysis. Crit Care 2015;19(1):245. DOI: 10.1186/s13054-015-0972-z.

19. Gibot S, Béné MC, Noel R, Massin F, Guy J, Cravoisy A, et al. Combination biomarkers to diagnose sepsis in the critically ill patient. Am J Respir Crit Care Med 2012;186(1):65–71. DOI: 10.1164/rccm.201201-0037OC.

20. Livaditi O, Kotanidou A, Psarra A, Dimopoulou I, Sotiropoulou C, Augustatou K, et al. Neutrophil CD64 expression and serum IL-8: Sensitive early markers of severity and outcome in sepsis. Cytokine 2006;36(5–6):283–290. DOI: 10.1016/j.cyto.2007.02.007.

21. Chen Q, Shi J, Fei A, Wang F, Pan S, Wang W. Neutrophil CD64 expression is a predictor of mortality for patients in the intensive care unit. Int J Clin Exp Pathol 2014;7(11):7806–7813. PMOD: 25550820.

22. Papadimitriou-Olivgeris M, Lekka K, Zisimopoulos K, Spiliopoulou I, Logothetis D, Theodorou G, et al. Role of CD64 expression on neutrophils in the diagnosis of sepsis and prediction of mortality in adult critically ill patients. Diagn Microbiol Infect Dis 2015;82(3):234–239. DOI: 10.1016/j.diagmicrobio.2015.03.022.

23. Huang Na, Chen J, Wei Yu, Liu Y, Yuan K, Chen J, et al. Multi-marker approach using C-reactive protein, procalcitonin, neutrophil CD64 index for the prognosis of sepsis in intensive care unit: A retrospective cohort study. BMC Infect Dis 2022;22;662. DOI: 10.1186/s12879-022-07650-6.

24. Dimoula A, Pradier O, Kassengera Z, Dalcomune D, Turkan H, Vincent JL. Serial Determinations of neutrophil CD64 expression for the diagnosis and monitoring of Sepsis in critically ill patients. Clin Infect Dis 2014;58(6):820–829 DOI: 10.1093/cid/cit936.

25. de Jong E, de Lange DW, Beishuizen A, van de Ven PM, Girbes AR, Huisman A, et al. Neutrophil CD64 expression as a longitudinal biomarker for severe disease and acute infection in critically ill patients. Int J Lab Hematol 2016;38(5):576–584. DOI: 10.1111/ijlh.12545.

26. Djordjevic D, Pejovic J, Surbatovic M, Jevdjic J, Radakovic S, Veljovic M, et al. Prognostic value and daily trend of interleukin-6, neutrophil CD64 expression, C-reactive protein and lipopolysaccharide-binding protein in critically ill patients: Reliable predictors of outcome or not? J Med Biochem 2015;34(4):431–439. DOI: 10.1515/jomb-2015-0002.

27. Ghosh PS, Singh H, Azim A, Agarwal V, Chaturvedi S, Saran S, et al. Correlation of neutrophil CD64 with clinical profile and outcome of sepsis patients during intensive care unit stay. Indian J Crit Care Med 2018;22(8):569–574. DOI: 10.4103/ijccm.

28. Ajmani S, Singh H, Chaturvedi S, Mishra R, Rai MK, Jain A, et al. Utility of neutrophil CD64 and serum TREM-1 in distinguishing bacterial infection from disease flare in SLE and ANCA-associated vasculitis. Clin Rheumatol 2019;38(4):997–1005. DOI: 10.1007/s10067-018-4334-5.

29. Baghdadi JD, Brook RH, Uslan DZ, Needleman J, Bell DS, Cunningham WE, et al. Association of a care bundle for early sepsis management with mortality among patients with hospital-onset or community-onset sepsis. JAMA Intern Med 2020;180(5):707–716. DOI: 10.1001/jamainternmed.2020.0183.