Original Article/Özgün Araştırma

doi: 10.54875/jarss.2023.87059

Prognostic Significance of Platelet and Mean Platelet Volume Kinetics for Ventilator-Associated Pneumonia

Platelet ve Ortalama Platelet Hacim Kinetiklerinin Ventilatör İlişkili Pnömoni İçin Prognostik Önemi

Omur Ilban[1], Fatih Simsek[2]

[1]Konya Meram State Hospital, Department of Intensive Care, Konya, Turkey
[2]University of Health Sciences, Gulhane Training and Research Hospital, Department of Anesthesiology and Reanimation, Ankara, Turkey

ABSTRACT

Objective: Platelets play an important role in cases of inflammation and infection, along with in the hemostatic process. The aim of study is to evaluate prognostic efficiency of platelet count (PC), mean platelet volume (MPV) and MPV/PC for mortality prediction in patients with ventilator-associated pneumonia (VAP).

Methods: A total of 150 VAP patients were divided into two groups as Survivors (n=98) and Non-survivors (n=52) according to intensive care unit mortality. The PC and MPV on days 1 and 4 of VAP were evaluated.

Results: Thirty (31%) patients in the survivors group and 35 (67%) patients in the non-survivor group had thrombocytopenia, respectively (p<0.05). The non-survivor group had higher MPV and a higher MPV/PC on the 1st and 4th days, while the survivor group had higher PC levels, respectively (p<0.05). Platelet count, MPV and MPV/PC had the highest AUC levels at day 4 (0.68, 0.80, 0.73, respectively). The prognostic values of day 4 MPV and MPV/PC measurements were similar (p=0.17). Day 4 PC were found to be negatively correlated with mortality (adjusted hazard ratio (aHR): 0.91, p=0.01). Day 4 MPV and MPV/PC values were the independent mortality predictors (aHR: 2.59, p<0.01 and aHR: 1.63, p=0.01). The survival probability of VAP patients with <9.25 fL of MPV day 4 was significantly higher than those with ≥9.25 fL (p=0.002, log-rank test)

Conclusion: In addition to thrombocytopenia, MPV and MPV/PC are useful predictors of poor outcomes. Changes in MPV responses may be a more useful prognostic assessment tool for VAP patients than PC.

Keywords: Immune response, mean platelet volume, platelets, prognosis, ventilator-associated pneumonia

ÖZ

Amaç: Trombositler inflamasyon ve infeksiyon durumlarında hemostatik süreçle birlikte önemli rol oynar. Çalışmanın amacı trombosit sayısı (PC), ortalama trombosit hacmi (MPV) ve MPV/PC'nin ventilatör ilişkili pnömonili (VAP) hastalarda mortalite tahmini için prognostik etkinliğini değerlendirmektir.

Yöntem: Toplam 150 VAP hastası, yoğun bakım ünitesi mortalitesine göre Survivors (n=98) ve Non-survivors (n=52) olarak iki gruba ayrıldı. Trombosit sayısı ve MPV, VAP'nin 1. ve 4. günlerinde değerlendirildi.

Bulgular: Survivors grubundaki 30 (%31) hastada ve Non-survivors grubundaki 35 (%67) hastada trombositopeni vardı (her ikisi için p<0,05). Non-survivors grubunda 1. ve 4. günlerde MPV ve MPV/PC daha yüksek bulunurken, Survivors grubu daha yüksek PC seviyelerine sahipti (her ikisi için p<0,05). Trombosit sayısı, MPV ve MPV/PC'nin AUC seviyeleri 4. günde en yüksekti (sırasıyla, 0,68, 0,80, 0,73). 4. gün MPV ve MPV/PC ölçümlerinin prognostik değerleri benzerdi (p=0,17). Dördüncü gün PC'nin mortalite ile negatif ilişkili olduğu bulundu (düzeltilmiş risk oranı (aHR): 0,91, p=0,01). Dördüncü gün MPV ve MPV/PC değerleri mortalitenin bağımsız göstergeleriydi (aHR: 2,59, p<0,01 and aHR: 1,63, p=0,01). Dördüncü gün MPV'si <9,25 fL olan VAP hastalarının hayatta kalma olasılığı, ≥9,25 fL olanlardan anlamlı olarak daha yüksekti (p=0,002, log-rank test).

Sonuç: Trombositopeniye ek olarak MPV ve MPV/PC, olumsuz sonuçların yararlı göstergeleridir. Ortalama trombosit hacmi yanıtlarındaki değişiklikler, VAP hastaları için PC'den daha yararlı bir prognostik değerlendirme seçeneği olabilir.

Anahtar sözcükler: İmmün yanıt, ortalama platelet hacmi, plateletler, prognoz, ventilatör ilişkili pnömoni

INTRODUCTION

In addition to hemostasis and thrombosis, platelets play critical roles in inflammation and the immune system (1,2). Similar to the leukocyte response, the platelet response in antimicrobial host defense contains antimicrobial peptides. Activated platelets clump together at the site of infection, forming a bridge between the antimicrobial peptides and the

Received/Geliş tarihi : 09.10.2022
Accepted/Kabul tarihi : 06.12.2022
Publication date : 30.01.2023

***Corresponding author:** Omur Ilban • droilban@gmail.com
Omur Ilban 0000-0001-8320-3150 / Fatih Simsek 0000-0002-8774-2861

Cite as: Ilban O, Fatih Simsek F. Prognostic significance of platelet and mean platelet volume kinetics for ventilator-associated pneumonia. JARSS 2023;31(1):28-36.

bacteria. Thrombocytopenia may be an early marker of community-acquired pneumonia (CAP) and sepsis. It is associated with an increase in the length of stay and mortality in hospital admission and treatment for thrombosis (3,4).

Mean platelet volume (MPV) is used as a marker to indicate the rate of platelet production in the bone marrow and its activation (5). Mean platelet volume has been considered an indicator of platelet function and activation because increased platelet volume and size point to a thrombotic and inflammatory state (6,7). Increased morbidity and mortality have been observed in cases with elevated MPV in pneumonia, ischemic stroke, cardiovascular diseases, and malignancy (8-11).

The term "ventilator-associated pneumonia" (VAP) refers to pneumonia that appears after endotracheal intubation, and more than 48 hours of mechanical ventilation (12). In the nosocomial setting, VAP continues to play a significant role in morbidity and mortality. Scores and biomarkers used until now have limited clinical utility for diagnosis and prognosis.

While platelet count (PC) and MPV changes have been linked to mortality in patients with sepsis and pneumonia, the number of studies on the prognostic significance of PC and MPV values in VAP patients is low. The study aims to assess the predictive value of the PC, MPV, and MPV/PC ratio as predictors of mortality in VAP patients.

MATERIAL and METHODS

Study Design

This retrospective, single-center study was conducted on adult patients with microbiologically-confirmed VAP in the 38-bed medical and surgical Intensive Care Unit (ICU) of Meram State Hospital. The study was approved by the ethics committee of Necmettin Erbakan University (approval number: 2022/3941) and performed between June 2020 and June 2022 following the Declaration of Helsinki. Because the study was retrospective, informed consent was not needed.

Inclusion and Exclusion Criteria

Patients were included in the study if they had a clinical suspicion of VAP as defined by the American Thoracic Society guidelines and a Clinical Pulmonary Infection Score (CPIS) above 6, if their tracheobronchial cultures had significant growth, and if they had no signs or symptoms of infection at the time of ICU admission (13,14).

Patients presenting underlying diseases that may affect platelet function or morphology, such as malignancy, chronic liver diseases, end-stage renal disease, primary hematological disorders, disseminated intravascular coagulation, severe thrombocytopenia (platelet count ≤ $20x10^9$ L^{-1}), receiving platelet transfusion, any extrapulmonary infection within the first 72 hours after pneumonia onset, isolation of fungal agents and organisms of unknown pathogenicity in tracheobronchial cultures, and patients who died in the 4 day observation period after the onset of VAP were excluded from the study.

The clinical characteristics of the patients were obtained from their medical records. These included age, sex, comorbidities [diabetes mellitus, renal dysfunction (estimated Glomerular Filtration Rate (eGFR) <60 mL min^{-1} 1.73 m^{-2}), cerebrovascular disease, cardiovascular disease, chronic lung disease], the severity of illness classification system Acute Physiology and Chronic Health Assessment (APACHE) II, CPIS/PaO_2/FiO_2 ratio, laboratory results (day 1 and day 4), vasopressor/inotrope use, and presence of septic shock data.

Definitions

A lower limit of 150,000 mm^{-3} platelet count was used to detect thrombocytopenia, and an MPV higher than 10.4 femtoliters (fL) was used to define increased MPV (15). Mean platelet volume to PC measurement was used to determine the MPV/PC ratio. According to Elsayed and Mohamed's research, the MPV/PC ratio is elevated if it is higher than 0.031 (16). The day of VAP onset was determined to be the day of the first positive tracheobronchial aspirate culture. Empirical antimicrobial therapy was initiated when the infectious disease specialist suspected VAP. Empirical antimicrobial therapy was defined as "appropriate" if the prescribed agents had in vitro activity against the infecting microorganisms. Disease severity was assessed by APACHE II, and CPIS scores were calculated from the collected data on admission (day 1). The same microorganisms isolated from VAP that cause bacteremia were accepted as positive blood cultures. The 30 day ICU mortality was defined as the mortality for any reason during hospitalization in the ICU within 30 days.

Study Protocol

During the study period, 150 consecutive VAP patients were included in the study. Patients were classified into two groups as survivors and non-survivors according to 30 day mortality. In patients with clinical suspicion of VAP and CPIS >6, the diagnosis of VAP was confirmed if there is a growth of $>10^4$ colony forming units (CFU) mL^{-1} in the conventional culture of the tracheobronchial aspirate (13,14). The PC and MPV measurements on days 1 and 4 of VAP were evaluated. A computerized analyzer (Sysmex XN-1000 Fluorescent Flow Cytometry, Norderstedt, Germany) was utilized at our institution to measure PC and MPV. Only the first VAP episode was studied in detail to avoid confounding effects on platelet markers related to individual normal variability or measured count variability of MPV.

Empirical antimicrobial therapy was administered in our hospital according to locally modified guidelines and was revised

based on antimicrobial test results (13). The pathogens isolated by the quantitative culture of the tracheobronchial aspirate were determined by standard microbiological methods. Blood and urine cultures were also taken at the same time to rule out other nosocomial infections. The VITEK 2 healthcare system (bioMérieux, France) was used for bacterial identification.

Statistical Analysis

The statistical analysis was performed using SPSS v.26.0 software (SPSS Inc., Chicago, IL, USA). Mean ± standard deviation and median (interquartile ranges) were used to present the continuous data, while integers and percentages were used to present the categorical data. Pearson's chi-squared or Fisher's exact tests were used in the analyses of categorical variables. Continuous variables with normal distributions were compared with the independent samples t-test, and those without normal distributions were compared with the Mann–Whitney U-test. Paired Samples t-test or Wilcoxon Signed-Rank test was used for repeated measurements. Receiver operating characteristic (ROC) analysis was performed to evaluate and compare the prognostic capacities of platelet indices found significant in the univariate analysis. The platelet biomarker with the largest area under the curve (AUC) of the ROC curve was examined by further analysis. The best discriminatory cut-off values were determined by the Youden index. Potential factors associated with mortality, including platelet indices, were evaluated with the multivariate Cox proportional hazard regression model, and the predictors were given as adjusted hazard ratios (aHRs) and 95% confidence intervals (CIs). The cumulative survival curve was defined with the Kaplan–Meier plot and compared with the log-rank test. All comparisons were two-sided, and $p<0.05$ was considered threshold for statistical significance.

The software G*Power version 3.1 was used to calculate the sample size required in our study. This analysis was performed based on the results of a similar study by Lee et al. (8). A sample size calculation was performed with a 95% confidence level ($p <0.05$), an effect size of 44%, and a power of 80% to check if there is a significant difference between the 1st and 4th day MPV changes of the two groups. Considering the 5% dropout rate, the sample size was calculated as 98 patients for group 1 and 52 patients for group 2 (150 patients in total).

RESULTS

A total of 227 patients with possible clinical VAP were recorded between the aforementioned dates. Among these 227 patients, pneumonia was microbiologically confirmed in 178 (78%) of them. In 18 patients, extrapulmonary infections other than pneumonia were found. Within four days of the onset of pneumonia, 10 patients died. The platelet indices of 150 patients with culture-confirmed VAP were reviewed.

Table I presents the clinical data of both patient groups. The median age of the patients was 65, and 76 (51%) patients were male. Cardiovascular diseases were the most prevalent comorbidity overall, accounting for 32 patients (21%). The APACHE II and CPIS scores of the non-survivor group were more likely to increase (22.1 vs. 25 and 7.2 vs. 7.4), while the PaO_2/FiO_2 ratio was less likely to decrease (279 vs. 246) ($p<0.05$). The number of days patients were intubated before developing VAP (6.9 vs. 5.3) and the number of days with length of ICU stay (17.1 vs. 14) were found to be statistically higher in the non-survivor group than in the survivor group ($p<0.05$).

All of the platelet indices compared in Table II demonstrated statistically significant differences between the groups. Thirty (31%) patients in the survivor group and 35 (67%) patients in the non-survivor group had thrombocytopenia ($p<0.05$). The non-survivor group had a higher MPV and a higher MPV/PC ratio on the 1st and 4th days, while the survivor group had higher PC levels ($p<0.05$). The decrease in platelet counts in the first 4 days of pneumonia was statistically significant in both the survivor and non-survivor groups ($p<0.01$). Although the increase in MPV values in the first 4 days of pneumonia was not significant in the survivor group ($p=0.18$), it was statistically significant in the non-survivor group ($p<0.01$). Similarly, while a significant change in MPV/PC ratios in the first 4 days of pneumonia in the survivor group was absent, the increase observed on the 4th day in the non-survivor group was significant ($p<0.01$) (data not shown). Unlike the survivor group, in all platelet indicators, significant differences were observed between days 1 and 4 in the non-survivor group. This indicates that dynamic changes in platelet response are significantly associated with mortality.

Since the laboratory parameters have a significant relationship with the mortality of VAP patients, the 1st and 4th day values of platelet indices were analyzed by ROC analysis as a predictive indicator of mortality. Mean platelet volume reached its highest AUC value of 0.80 on day 4. Platelet counts had the lowest 1st and 4th day AUC values among all parameters (0.65 and 0.68, respectively). Mean platelet volume on day 4, with the best sensitivity (79%) and a cut-off value of 9.25 fL, and platelet count on day 4, with the best specificity (79%) and a cut-off value of 192.5, were the prognostic markers of 30 day mortality.

Platelet indices, which have relatively low AUC (<0.80) in single measurements and do not have sufficient capacity to predict mortality, were examined in terms of the best prognostic value reached with pairwise comparisons and combinations. When ROC curves were compared pairwise with MPV on day

Table I. Clinical Characteristics of Patients with Ventilator Associated Pneumonia

Variables	Total (n=150)	Survivors (n=98)	Non-Survivors (n=52)	p
Age (year)	65 (58-74)	64.5 (58-72.3)	68 (57.5-77)	0.08
Male	76 (51)	48 (49)	28 (54)	0.57
Comorbidities				
Diabetes mellitus	28 (19)	16(16)	12 (23)	0.31
Renal dysfunction	22 (15)	13 (13)	9 (17)	0.51
Cerebrovascular disease	14 (9)	10 (10)	4 (8)	0.77
Cardiovascular disease	32 (21)	20 (20)	12 (23)	0.7
Chronic lung disease	22 (15)	12 (12)	10 (19)	0.25
APACHE II	23 (21-27)	22.1 (20.2-26.3)	25 (22.1-27.7)	<0.01*
CPIS	7.3 ± 0.2	7.2 ± 0.2	7.4 ± 0.3	0.01*
PaO_2/FiO_2 ratio	263 (184-304)	279 (185-311)	246 (174-284)	0.02*
Vasopressor/inotrope use	55 (37)	31(32)	24 (46)	0.08
Septic shock	38 (25)	22 (22)	16 (31)	0.27
Positive blood culture	16 (11)	7 (7)	9 (17)	0.06
Microbiological profile				
Polimicrobial	26 (17)	15 (15)	11 (21)	0.37
Gram negative rods	96 (64)	61 (62)	35 (67)	0.54
Gram positive cocci	28 (19)	22 (22)	6 (12)	0.1
Inappropriate empirical theraphy	49 (33)	27 (28)	22 (42)	0,07
Duration of intubation prior to VAP (days)	5.9 ± 2.2	5.3 ± 2.1	6.9 ± 2.1	<0.01*
Length of ICU stay (days)	15.1 ± 7.1	14 ± 7	17.1 ± 6.9	0.01*

Data shown as mean ± standard deviation, median (interquartile ranges) or n (%). * Survivors vs Non-survivors group (p <0.05). **APACHE II:** Acute physiological and chronic health evaluation, **CPIS:** Clinical pulmonary infection score, **PaO_2/FiO_2:** Ratio of arterial oxygen concentration to the fraction of inspired oxygen, **VAP:** Ventilator associated pneumonia, **ICU:** Intensive care unit.

Table II. Platelet Indices of Patients Among Survivors and Non-Survivors

Variables	Total (n=150)	Survivors (n=98)	Non-Survivors (n=52)	p
Incidence of thrombocytopenia	65 (43)	30 (31)	35 (67)	<0.01*
Incidence of increased MPV	41 (27)	18 (18)	23 (44)	<0.01*
Platelet day 1	235 (120-280)	239 (204-280)	149 (95-297)	<0.01*
Platelet day 4	218 (126-295)	221 (210-285)	138 (91-287)	<0.01*
MPV day 1	8.83 ± 1.1	8.54 ± 1	9.38 ± 1.2	<0.01*
MPV day 4	9.16 ± 1.3	8.68 ± 1.2	10.07 ± 1.2	<0.01*
MPV/PC ratio day 1	0.05 ± 0.03	0.04 ± 0.03	0.07 ± 0.04	<0.01*
MPV/PC ratio day 4	0.06 ± 0.04	0.05 ± 0.03	0.08 ± 0.05	<0.01*

Data shown as mean ± standard deviation, median (interquartile ranges) or n (%). *Survivors vs Non-survivors group (p<0.05). **MPV:** Mean platelet volume, **MPV/PC:** Ratio of mean platelet volume to platelet count.

4 to predict mortality, the differences in the AUC were statistically significant except for the MPV/PC ratio on day 4 (p<0.05 for both). Therefore, MPV and MPV/PC ratio values on day 4 had similar prognostic values in evaluating mortality risk in VAP patients (p=0.17). The combined evaluation of MPV values higher than 9.25 fL on day 4 with MPV/PC ratio values higher than 0.05 on day 4, did not improve specificity (71%), but it did increase sensitivity (88%) and AUC (0.84). However, no significant difference was observed between the prognostic values of both parameters (the difference between areas:

Table III. Performance of Platelet Indices for Predicting 30-Day Mortality

Variables	Cut-off	AUC (95% CI)	Sensitivity (%)	Specificity (%)	PPV	NPV	p
PC day 1	208.5	0.65 (0.54-0.75)	73	76	61.3	84.1	0.01*
PC day 4	192.5	0.68 (0.59-0.79)	73	79	64.4	84.6	0.01*
MPV day 1	8.85	0.71 (0.62-0.80)	67	68	52.2	79.5	0.04*
MPV day 4	9.25	0.80 (0.73-0.88)	79	78	66.1	87.5	-
MPV/PC ratio day 1	0.04	0.66 (0.58-0.77)	71	65	44.3	79	0.01*
MPV/PC ratio day 4	0.05	0.73 (0.66-0.80)	72	71	56.1	82.1	0.17
MPV 4 + MPV/PC 4		0.84 (0.77-0.89)	88	71	62.2	92.1	0.08

Receiver operating characteristic plot analysis of platelet indices with respect to prediction of ICU mortality. The p values correspond to the difference between the AUC of the parameters and the AUC of MPV day 4. Data are presented as %, unless otherwise stated. **AUC:** Area under the receiver-operating-characteristic curve, **CI:** Confidence interval, **PPV:** Positive predictive value, **NPV:** Negative predictive value, **PC:** Platelet count, **MPV:** Mean platelet volume, **MPV/PC:** Ratio of mean platelet volume to platelet count.

0.04; SE: 0.02; 95% CI, –0.00 to 0.07; z = 1.77; p=0.08) (Table III).

Cox hazard regression was used to analyze platelet disorder and mortality. Day 4 PCs were negatively correlated with mortality in the multivariate analysis (adjusted for age, sex, APACHE II score, and renal dysfunction) using the MPV/PC ratio as the categorical variable (aHR: 0.91, 95% CI: 0.81-0.98, p=0.01). In terms of mortality, MPV day 4 values and MPV/PC ratio day 4 values were independent predictors (aHR: 2.59, 95% CI: 1.64-4.10, p<0.01 and aHR: 1.63, 95% CI: 1.05-2.69, p=0.01, respectively) (Figure 1).

The 30-day mortality from the onset of pneumonia was analyzed with the Kaplan–Meier curve concerning the cut-off value of the day 4 MPV values extracted from our data. When the 9.25 fL cut-off value for day 4 MPV was used, the probability of survival in VAP patients with MPV smaller than 9.25 fL was significantly higher than those with higher than 9.25 fL MPV (p=0.002, log-rank test) (Figure 2).

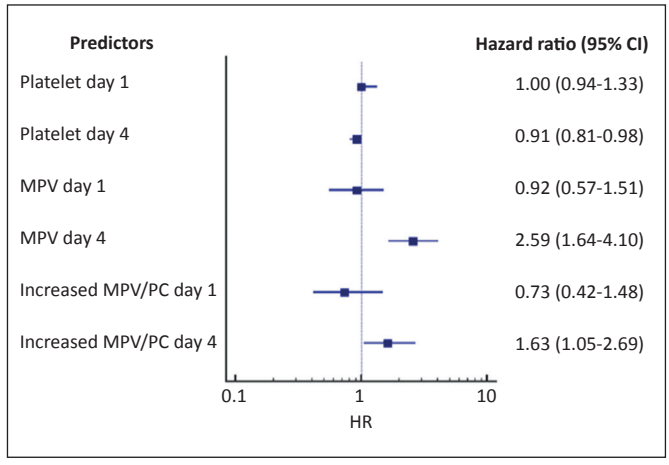

Figure 1. Platelet indices that were possibly associated with low survival. Hazard ratio was calculated using a Cox proportional-hazards model adjusted by age, gender, APACHE II score and renal dysfunction. **CI:** Confidence interval, **MPV:** Mean platelet volume, **MPV/PC:** Ratio of mean platelet volume to platelet count.

DISCUSSION

This study showed that platelet and MPV responses were significantly correlated with mortality in VAP patients. Compared to PC, MPV was a better indicator of negative outcomes in pneumonia that developed during mechanical ventilation.

Platelets play a role in inflammatory and immune complications in sepsis. While interactions with immune cells may be beneficial in maintaining hemostasis and limiting infection, ongoing platelet interactions may have harmful effects by overstimulating the immune system and resulting in inflammatory damage (1). In experimental models of pneumosepsis, platelet activation was associated with sepsis caused by *Klebsiella pneumoniae* and *Staphylococcus aureus* (17,18). In these studies, thrombocytopenia was associated with an

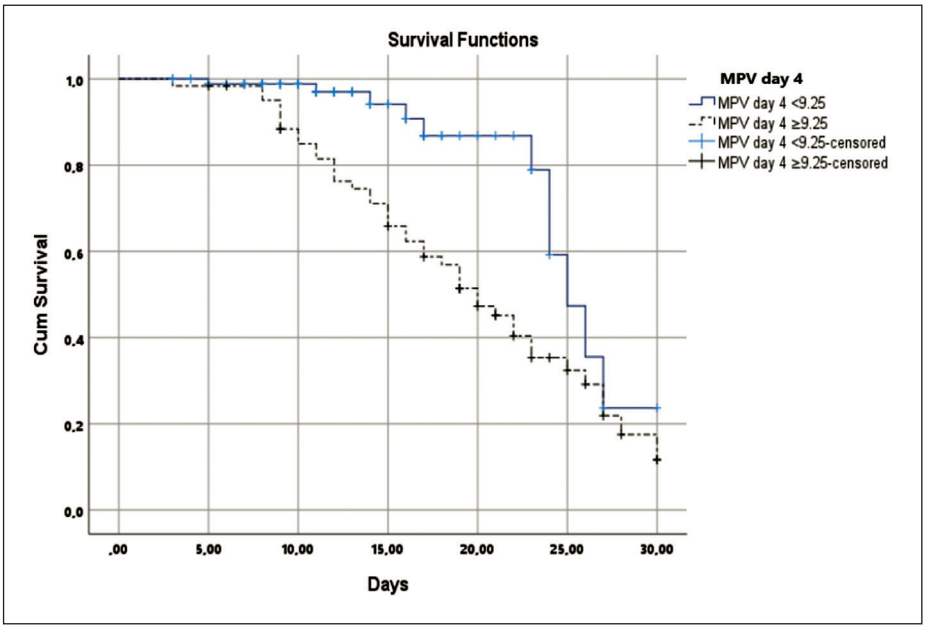

Figure 2. Kaplan-Meier curves of the ventilator associated pneumonia patients stratified by MPV day 4. **MPV:** Mean platelet volume.

increase in pulmonary bacterial load and systemic infection. Similarly, thrombocytopenia seen in patients with CAP and sepsis was correlated with low survival (3,4).

Cardiovascular events that occur during infection may cause death in pneumonia patients and platelet activation has an important role in these events (19). In a recent prospective study, Tunjungputri et al. found that the pb1B gene expression by *Streptococcus pneumoniae* is associated with increased platelet activation and 30-day mortality (20). Consistent with the previously mentioned studies, in this study, the reduction in the PC of VAP patients may be associated with negative outcomes, and an increase in disease severity may contribute to a higher risk of mortality in these patients.

A mortality risk factor for the disease is thrombocytopenia which appears at the beginning of the illness or during the ICU stay (3,4). While PC remained above the threshold for thrombocytopenia, Sezgi et al. hypothesized that the decrease seen in patients with normal PC at ICU admission during the follow-up period was a poor indicator of the disease process (21). From day 1 to day 4, the non-survivor group's median PC levels in our study showed a clear decrease. The non-survivors' values dropped below the thrombocytopenia threshold at the same time for both measurements. However, only day 4 PC was independently associated with mortality according to the results of the multivariate regression analysis. According to our findings, compatible with those of a previous study, a decrease in PC during the disease may be a more significant prognostic factor than the presence of thrombocytopenia.

Mean platelet volume measurements were shown to be associated with mortality in severe infections. In their study based on a single MPV measurement, Golcuk et al. showed that MPV level was a predictor of mortality in CAP, and its predictive value increased when combined with the CURB-65 score (22). This score is composed of 5 variables with a weight of 1 point in each of them: new onset confusion, urea level higher than 7 mmol L^{-1}, Respiratory rate higher than 30 min^{-1}, systolic Blood pressure lower than 90 mmHg, and/or diastolic blood pressure lower and equal to 60 mmHg for patients aged above 65. In their study of patients with septic shock, Gao et al. found that MPV had the highest prognostic value for in-hospital mortality among platelet indices (23). In the study of Gorelik et al. investigating the clinical features and prognostic value of MPV changes in CAP patients, an increase in MPV values was correlated with an increase in pneumonia severity and mortality (24). However, when the baseline MPV measurements were analyzed using cut-off values similar to those of Golcuk et al., there was no significant difference in in-hospital and long-term mortality. Therefore, it has been suggested that a single MPV measurement may be an indicator of disease severity and comorbidities rather than a predictor of mortality. In this study, in the time-dependent mortality analysis in which MPV values on the first day of pneumonia were measured, MPV day 4 values were positively associated with mortality, although day 1 MPV values were not associated with negative outcomes. These results parallel to previous studies suggest that repeated MPV measurements can significantly contribute to risk assessment and prognostic performance.

A high MPV is related to advanced age, sex, renal dysfunction, and disease severity (24). The possible mechanisms for the increase in MPV may be explained by severe inflammation,

increasing thrombopoiesis, inflammatory cytokines, and the increased expression of younger large platelets, as well as higher thrombocyte consumption in peripheral tissues and spleen (25). Mild and moderate renal dysfunction, which we described as an estimated glomerular filtration rate lower than mL min^{-1} 1.73 m^{-2} at the onset of pneumonia, may be associated with an increase in MPV, although a risk factor that may confuse was the exclusion of renal replacement therapy in our study. As previously stated, the robustness of the prognostic potential of MPV in VAP patients has been demonstrated by including confounding factors, including age, sex, APACHE II score, and renal dysfunction, associated with changes in MPV levels in the regression analysis.

Although the underlying mechanisms of the relationship between MPV increase and negative outcomes are not known enough, the increase in platelet activation may be the main mechanism. Mean platelet volume is an indicator of platelet activation as measured by platelet size. Larger and reactive platelets with increased MPV are associated with inflammatory and thrombotic conditions (26). More activated larger platelets express more procoagulant substances, including intracellular thromboxane A2 (TXA2) and surface proteins such as p-selectin and glycoprotein IIIa. Therefore, the prolongation of vasoconstriction, the increase in platelet aggregation, and the development of endothelial dysfunction may increase the risk of cardiovascular thrombosis and mortality in patients with increased MPV (27). At the same time, TXA2 can activate the pulmonary vascular endothelium, which is an important component of acute lung injury due to severe infection. Moreover, increased platelet activation causes a greater inflammatory response by increasing the release of immunomodulatory ligands and cytokines from platelets (28). The abovementioned adverse effects due to continued activation of larger platelets may be a potential mechanism related to the increased mortality in VAP patients.

Platelet volume may be a better indicator than PC in the evaluation of platelet function (29). Aydemir et al. investigated PC and MPV changes and infection-specific responses in sepsis patients. The decreases in platelet counts of gram-positive and gram-negative septic patients were associated with the simultaneous increase in MPV (15). In the study conducted by Sezgi et al. in the respiratory ICU, while MPV levels increased, the decrease in PC was associated with mortality (21). Last, Cho et al. examined the MPV/PC ratio in terms of mortality risk in CAP patients. The MPV/PC ratio presented a higher prognostic value than MPV or PC alone, and it was also equivalent to procalcitonin (30). However, the results of this study showed a similar prognostic value between the MPV/PC ratio and MPV in VAP patients and showed that MPV/PC is a weaker predictor of mortality than MPV.

In this study, the prognostic capacity of platelet indices in VAP patients was examined. Mean platelet volume on day 4 had the highest AUC of 0.80 and the highest sensitivity at 79%, while PC on day 4 had the highest specificity at 79%. While MPV day 4 showed a good discriminative capacity for mortality, other platelet parameters had adequate discriminative capacity with AUC <0.8. At the same time, the prognostic value of MPV day 4 was higher than that of all platelet parameters except the MPV/PC ratio on day 4. In addition, the combination of the two platelet indices with the best discriminative capacity, MPV on day 4 and MPV/PC ratio day 4, reached a higher AUC of 0.84 than their values alone, but its prognostic value did not change. Therefore, both platelet parameters may be useful indicators for estimating mortality in patients with VAP.

Several limitations are included in this study. First, it is limited to being single-center and retrospective. Second, due to the retrospective design of our study, dynamic changes in platelet counts and MPV measurements in response to treatment could not be evaluated. Third, due to the lack of data on other inflammatory markers, such as procalcitonin and C-reactive protein, the prognostic values of platelet indices and these markers could not be compared. Fourth, the possible specific platelet response to different pathogens causing VAP has not been evaluated. Finally, the majority of VAP patients recruited were elderly, had higher disease severity, and had multiple underlying diseases. Therefore, unmeasured differences and residual confounders between groups may affect the results observed.

CONCLUSION

In addition to the risk associated with thrombocytopenia in the early phase of VAP, MPV, and MPV/PC ratio measurements are useful predictors of negative outcomes. Mean platelet volume may be a better predictor of platelet function and activation than PC. The evaluation of VAP patients' platelet reactions may be useful in determining the adequacy of diagnosis and treatment, as well as the potential complications.

AUTHOR CONTRIBUTIONS

Conception or design of the work: OI, FS
Data collection: OI
Data analysis and interpretation: OI, FS
Drafting the article: OI, FS
Critical revision of the article: OI, FS
Other (study supervision, fundings, materials, etc): OI, FS
All authors (OI, FS) reviewed the results and approved the final version of the manuscript.

REFERENCES

1. Morrell CN, Aggrey AA, Chapman LM, Modjeski KL. Emerging roles for platelets as immune and inflammatory cells. Blood 2014;123(18):2759-67.
2. Semple JW, Italiano JE Jr, Freedman J. Platelets and the immune continuum. Nat Rev Immunol 2011;11(4):264-74.
3. Mirsaeidi M, Peyrani P, Aliberti S, et al. Thrombocytopenia and thrombocytosis at time of hospitalization predict mortality in patients with community-acquired pneumonia. Chest 2010;137(2):416-20.
4. Vanderschueren S, De Weerdt A, Malbrain M, et al. Thrombocytopenia and prognosis in intensive care. Crit Care Med 2000;28(6):1871-6.
5. Ekiz F, Yuksel O, Kocak E, et al. Mean platelet volume as a fibrosis marker in patients with chronic hepatitis B. J Clin Lab Anal 2011;25(3):162-5.
6. Shah B, Valdes V, Nardi MA, Hu L, Schrem E, Berger JS. Mean platelet volume reproducibility and association with platelet activity and anti-platelet therapy. Platelets 2014;25(3):188-92.
7. Thompson CB, Eaton KA, Princiotta SM, Rushin CA, Valeri CR. Size dependent platelet subpopulations: relationship of platelet volume to ultrastructure, enzymatic activity, and function. Br J Haematol 1982;50(3):509-19.
8. Lee JH, Park MA, Han SW, Hwang JJ, Park SH, Park SY. An increase in mean platelet volume during admission can predict the prognoses of patients with pneumonia in the intensive care unit: A retrospective study. PLoS One 2018;13(12):e0208715.
9. Arévalo-Lorido JC, Carretero-Gómez J, Álvarez-Oliva A, Gutiérrez-Montaño C, Fernández-Recio JM, Najarro-Diez F. Mean platelet volume in acute phase of ischemic stroke, as predictor of mortality and functional outcome after 1 year. J Stroke Cerebrovasc Dis 2013;22(4):297-303.
10. Wasilewski J, Desperak P, Hawranek M, et al. Prognostic implications of mean platelet volume on short and long-term outcomes among patients with non-ST-segment elevation myocardial infarction treated with percutaneous coronary intervention: A single-center large observational study. Platelets 2016;27(5):452-8.
11. Pyo JS, Sohn JH, Kang G. Diagnostic and prognostic roles of the mean platelet volume in malignant tumors: A systemic review and meta-analysis. Platelets 2016;27(8):722-8.
12. Vincent JL, Bihari DJ, Suter PM, et al. The prevalence of nosocomial infection in intensive care units in Europe. Results of the European Prevalence of Infection in Intensive Care (EPIC) Study. EPIC International Advisory Committee. JAMA 1995;274(8):639-44.
13. Kalil AC, Metersky ML, Klompas M, et al. Management of adults with hospital-acquired and ventilator-associated pneumonia: 2016 Clinical Practice Guidelines by the Infectious Diseases Society of America and the American Thoracic Society. Clin Infect Dis 2016;63(5):e61-e111.
14. Luna CM, Blanzaco D, Niederman MS, et al. Resolution of ventilator associated pneumonia: prospective evaluation of the clinical pulmonary infection score as an early clinical predictor of outcome. Crit Care Med 2003;31(3):676-82.
15. Aydemir H, Piskin N, Akduman D, Kokturk F, Aktas E. Platelet and mean platelet volume kinetics in adult patients with sepsis. Platelets 2015;26(4):331-5.
16. Elsayed AM, Mohamed GA. Mean platelet volume and mean platelet volume/ platelet count ratio as a risk stratification tool in the assessment of severity of acute ischemic stroke. Alex J Med 2017;53(1):67-70.
17. de Stoppelaar SF, Van't Veer C, Roelofs JJ, et al. Platelet and endothelial cell P-selectin are required for host defense against Klebsiella pneumoniae-induced pneumosepsis. J Thromb Haemost 2015;13(6):1128-38.
18. Youssefian T, Drouin A, Masse JM, Guichard J, Cramer EM. Host defense role of platelets: Engulfment of HIV and Staphylococcus aureus occurs in a specific subcellular compartment and is enhanced by platelet activation. Blood 2002;99(11):4021-9.
19. Rae N, Finch S, Chalmers JD. Cardiovascular disease as a complication of community-acquired pneumonia. Curr Opin Pulm Med 2016;22(3):212-8.
20. Tunjungputri RN, van de Heijden W, Urbanus RT, de Groot PG, van der Ven A, de Mast Q. Higher platelet reactivity and platelet-monocyte complex formation in Gram-positive sepsis compared to Gram-negative sepsis. Platelets 2017;28(6):595-601.
21. Sezgi C, Taylan M, Kaya H, et al. Alterations in platelet count and mean platelet volume as predictors of patient outcome in the respiratory intensive care unit. Clin Respir J 2015;9(4):403-8.
22. Golcuk Y, Golcuk B, Bilge A, Irik M, Dikmen O. Combination of mean platelet volume and the CURB-65 score better predicts 28-day mortality in patients with community-acquired pneumonia. Am J Emerg Med 2015;33(5):648-52.
23. Gao Y, Li Y, Yu X, et al. The impact of various platelet indices as prognostic markers of septic shock. PLoS One 2014;9(8):e103761.
24. Gorelik O, Tzur I, Barchel D, et al. A rise in mean platelet volume during hospitalization for community-acquired pneumonia predicts poor prognosis: A retrospective observational cohort study. BMC Pulm Med 2017;17(1):137.
25. Noris P, Melazzini F, Balduini CL. New roles for mean platelet volume measurement in the clinical practice? Platelets 2016;27(7):607-12.
26. Kim CH, Kim SJ, Lee MJ, et al. An increase in mean platelet volume from baseline is associated with mortality in patients with severe sepsis or septic shock. PLoS One 2015;10(3):e0119437.
27. Chu SG, Becker RC, Berger PB, et al. Mean platelet volume as a predictor of cardiovascular risk: A systematic review and meta-analysis. J Thromb Haemost 2010;8(1):148-56.

28. Kerris EWJ, Hoptay C, Calderon T, Freishtat RJ. Platelets and platelet extracellular vesicles in hemostasis and sepsis. J Investig Med 2020;68(4):813-20.
29. Tajarernmuang P, Phrommintikul A, Limsukon A, Pothirat C, Chittawatanarat K. The role of mean platelet volume as a predictor of mortality in critically ill patients: A systematic review and meta-analysis. Crit Care Res Pract 2016;2016:4370834.
30. Cho J, Lee S, Uh Y, Lee JH. Usefulness of mean platelet volume to platelet count ratio for predicting the risk of mortality in community-acquired pneumonia. Arch Med Sci 2020;16(6):1327-35.