



# Diagnostic Role of Platelet to Lymphocyte Ratio and Platelet Parameters in COVID-19 Disease

## ARTICLE INFO

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## ABSTRACT

**Backgrounds:** This study aimed to analyze the applicability of platelet parameters in assessing the severity of COVID-19 disease.

**Materials & Methods:** Patients with RT-PCR confirmed COVID-19 in the Pathology department of a tertiary care hospital in south India from June to December 2020 were included in this study. Clinical details and laboratory parameters of these patients were obtained. The difference between the studied variables in two groups was assessed using independent t-test. The optimum cut-off value of platelet to lymphocyte ratio (PLR) to differentiate between the tested groups was estimated using ROC (receiver operator curve) analysis.

**Findings:** This study was conducted on 218 COVID-19 patients, of whom 17.9% showed thrombocytopenia at the time of admission. Among the hematological parameters, PLR, absolute lymphocyte count (ALC), platelet distribution width (PDW), D-dimer, and erythrocyte sedimentation rate (ESR) were significantly different between the ICU (intensive care unit) and non-ICU groups. Increased PLR values were associated with the disease severity.

**Conclusion:** PLR could be used as an additional biomarker in assessing the severity of COVID-19 disease, and a cut-off value of 210.27 is optimal to differentiate severe COVID-19 disease from its mild and moderate forms with 79% specificity.

**Keywords:** Biomarker, COVID-19, Intensive care unit.

## CITATION LINKS

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## Introduction

Coronavirus is a RNA virus that belongs to the family *Coronaviridae*. The genera that cause human infections are predominantly alpha and beta. These groups of viruses mainly cause the common cold, whereas Middle East respiratory syndrome coronavirus (MERS-CoV) and severe acute respiratory distress syndrome coronavirus (SARS-CoV) mainly cause severe acute respiratory syndrome in humans. The newly discovered beta coronavirus variant belongs to the same genus as SARS-CoV and MERS-CoV but differs in its genetic characteristics. In February 2020, the World Health Organization (WHO) named the infection caused by the novel coronavirus as COVID-19 [1]. The novel virus was later named by the International Committee on Virus Taxonomy as severe acute respiratory distress syndrome–coronavirus-2 (SARS-CoV-2) [2].

The clinical manifestations of this disease range from an asymptomatic illness to severe acute respiratory failure or multi-organ failure, leading to death. Although the mechanism causing severity is not known, co-morbid conditions such as old age, diabetes, hypertension, and pulmonary and cardiovascular diseases are associated with the disease severity. Unusual symptoms of this disease include abdominal pain, headache, palpitations, and chest pain. Decreased lymphocyte and platelet counts and eosinopenia are among the alterations observed in hematological parameters of COVID-19 patients [3, 4]. Changes in coagulation parameters like prolonged activated partial thromboplastin time (aPTT), increased D-dimer values, and raised erythrocyte sedimentation rate (ESR) have also been reported in these patients [5]. These are evidenced by hypercoagulable states and micro-thrombi formation in acutely ill patients as described in a previous study [6].

A complete blood count (CBC) performed on all COVID-19 patients at the time of ad-

mission includes absolute lymphocyte count (ALC), platelet count (PLT), and platelet parameters such as mean platelet volume (MPV) and platelet distribution width (PDW). The latter two are useful in predicting the cause of thrombocytopenia and also reflect platelet activation as proven in many studies [7]. CBC data such as platelet count and absolute lymphocyte count could be used to calculate the platelet to lymphocyte ratio (PLR). Studies has revealed the role of these parameters in predicting the severity of COVID-19 disease [8-10].

**Objectives:** This study aimed to analyze the applicability of platelet parameters in assessing the severity of COVID-19 disease. Thus, platelet parameters and PLR were compared between two groups of COVID-19 patients: those requiring intensive care (ICU patients) and those not requiring intensive care (non-ICU patients).

## Materials and Methods

This prospective observational study was conducted in the Pathology department of a tertiary care hospital in India from June to December 2020. All RT-PCR (reverse transcription–polymerase chain reaction)-confirmed COVID-19 patients with available baseline hematological parameters were included in this study. Ethical approval was obtained from the Institutional Ethics Committee. The clinical data of patients obtained from their hospital medical records were assessed to stratify the disease severity.

The clinical management protocol as per Ministry of Health and Family Welfare (MoHFW) guidelines [11] was followed by the institutional COVID-19 task force team. Patients were clinically stratified by disease severity into mild, moderate, and severe categories based on MoHFW guidelines [11]. Patients with signs and symptoms such as fever, cough, sore throat, vomiting, diarrhea, etc. but without shortness of breath, dyspnea,

or abnormal chest imaging were considered to have mild COVID-19. Patients with lower respiratory disease diagnosed by clinical assessments or imaging and an oxygen saturation level ( $\text{SpO}_2$ )  $\geq 94\%$  were considered to have moderately severe COVID-19. Those with  $\text{SpO}_2 < 94\%$ , ratio of partial pressure arterial oxygen to fraction of inspired oxygen ( $\text{PaO}_2/\text{FiO}_2$ )  $< 300$  mmHg, respiratory rate  $> 30$  breath/min, or lung infiltrates  $> 50\%$  were considered as severely ill. Based on their clinical conditions, patients were divided into two groups: those with severe disease requiring intensive care (ICU patients) and those with mild / moderate disease not requiring intensive care (non-ICU patients). The clinical data of these patients were obtained from the hospital information system and recorded. The recorded parameters included absolute lymphocyte count (ALC), platelet count (PLT), mean platelet volume (MPV), platelet distribution width (PDW), D-dimer, prothrombin time (PT), and activated partial thromboplastin time (aPTT). Erythrocyte sedimentation rate (ESR) and interleukin-6 (IL-6) values were also recorded whenever available.

Platelet trend was assessed by calculating the serial platelet count values of ICU patients during their hospital stay.

PLR was calculated using ALC and PLT counts based on the following formula:  $\text{PLR} = \text{ALC}/\text{PLT} \times 100$ .

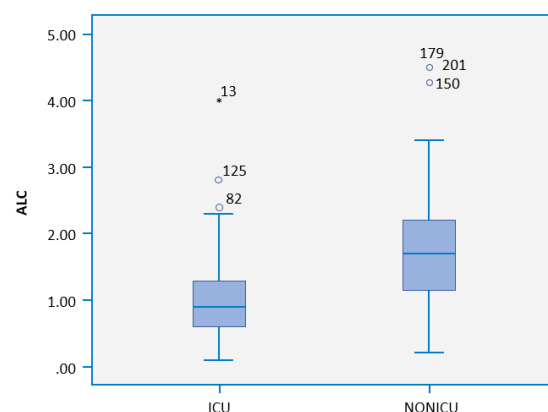
**Data analysis:** Data were recorded and analyzed using Microsoft Excel and SPSS software Version 24.0. The studied variables were expressed as mean and standard deviation. The difference in parameters between the two groups was assessed by independent student's t-test. A  $p$ -value of  $< .05$  was considered statistically significant. Receiver operator curve (ROC) analysis was done to determine the cut-off value of PLR to differentiate between the ICU and non-ICU groups. If the area under the curve is closer

to one, it indicates maximum sensitivity and specificity.

## Findings

A total of 218 RT-PCR confirmed COVID-19 patients were included in the study. Among them, 145 (66.5%) patients were treated in the ICU, and 73 (33.5%) patients were treated in other wards (non-ICU group). Table 1 compares the hematological and biochemical parameters between the two groups.

**Comparison of ALC between the ICU and non-ICU groups:** The mean ALC at the time of admission in the non-ICU and ICU groups was  $1.8$  and  $0.99 \times 10^3$  cells/ $\text{mm}^3$ , respectively. At the time of admission, 78% of ICU patients and 4% of non-ICU patients had lymphopenia (ALC less than  $1 \times 10^3$  cells/ $\text{mm}^3$ ). These results showed a statistically significant difference between the ICU and non-ICU groups (Figure 1).

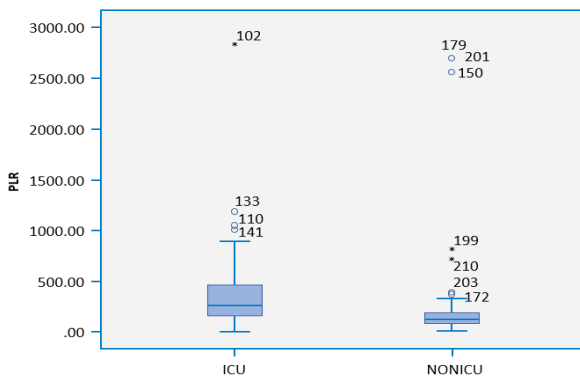


**Figure 1:** Comparison of ALC values between the ICU and non-ICU groups in the present study. At the time of admission, 78% of ICU patients and 4% of non-ICU patients had lymphopenia with  $\text{ALC} < 1 \times 10^3$  cells/ $\text{mm}^3$ .

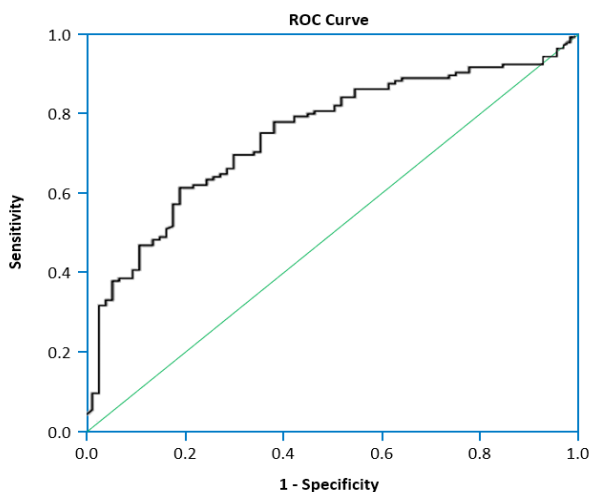
**Comparison of PLT count and extended platelet parameters between the ICU and non-ICU groups:** PLT count: At the time of admission, the mean PLT count in the non-ICU and ICU groups was 234.71 and 242.96 cells/ $\text{mm}^3$ , respectively. A total of 39 pa-

tients (17.9%) showed thrombocytopenia at the time of admission, among which 28% were in the ICU group, and 11% were in the non-ICU group. Hence, the platelet count showed no statistically significant difference between the two groups.

a. PLR: The mean PLR in the non-ICU and ICU groups was 166.8 and 340, respectively. Thus, the PLR value was significantly higher in the ICU group (Figure 2). The peak PLR value observed in the ICU group during the hospital stay was 1193. ROC analysis showed an optimum cut-off value of 210.27 for PLR to determine the disease severity with a sensitivity of 61% and specificity of 79% (Figure 3).



**Figure 2)** Comparison of PLR between ICU and non-ICU patients in the present study. ICU patients had significantly higher PLR values.

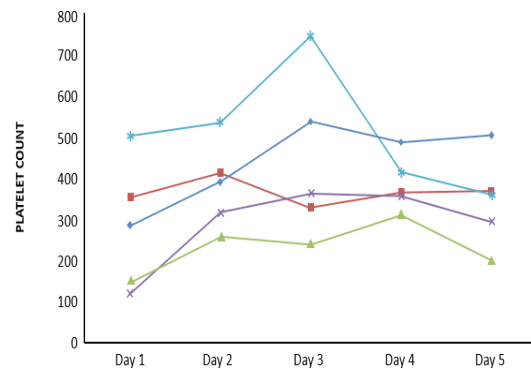


**Figure 3)** ROC analysis of PLR value to differentiate between ICU and non-ICU patients in the present study. Optimum cut-off value for severity was 210.27 with a sensitivity of 61% and specificity of 79%.

b. MPV & PDW: As shown in Table 1, among the mean MPV and PDW, PDW showed a statistically significant difference between the ICU and non-ICU groups ( $p < .001$ ).

c. In this study, inflammation-associated biomarkers such as D-dimer, PT, aPTT, ESR, and IL-6 were also analysed. PT and aPTT did not show much difference between the two groups, but IL-6 value was higher in the ICU group. D-dimer and ESR showed a statistically significant difference between the two groups.

**Analysis of PLT trend:** The variation in PLT count of ICU patients during their hospital stay showed a roller-coaster pattern with an initial decrease followed by an increase and then a decrease. The PLT trend in ICU patients is illustrated in Figure 4.



**Figure 4:** Variation in platelet count of ICU patients during their hospital stay in the present study

a. Peak PLT value: The peak PLT value recorded during the hospital stay was 594 and 818x10<sup>3</sup> cells/mm<sup>3</sup> in the non-ICU and ICU groups, respectively.

**Peripheral smear study of platelets:**

Peripheral smear test was done to confirm the platelet counts and observe the morphology of platelets. Interestingly, platelet anisocytosis, immature megakaryocytes, and platelet phagocytosis by activated lymphocytes were found in few cases with thrombocytopenia.

**Table 1:** Comparison of hematological and biochemical parameters between the ICU and non-ICU groups in the present study

S. No	Parameters	ICU (N=145)		Non -ICU (N=73)		P Value
		Mean	SD	Mean	SD	
1.	ALC (X1000)	0.99	0.57	1.8068	0.89	< .05*
2.	PLT count (x1000)	242.96	111.17	234.71	95.59	.589
3.	PLR	339.70	310.57	166.87	130.47	< .05*
4.	MPV	8.92	5.46	8.58	1.00	.594
5.	PDW	17.42	0.71	16.85	0.77	< .05*
6.	D-dimer	2.53	4.216	0.70	0.96	< .05*
7.	PT	12.40	2.17	13.78	6.40	.102
8.	APTT	25.98	6.19	28.90	3.06	.171
9.	ESR	43.19	25.80	24.16	23.82	< .05*
10.	IL-6	138.67	381.05	53.99	193.09	.096

\*indicates significance at  $p < .05$ .

## Discussion

According to immunological research, high amounts of pro-inflammatory cytokines, often known as a cytokine storm, have been identified as a feature of severely ill COVID-19 patients. Multiple organ dysfunction syndrome and acute respiratory distress syndrome, which contribute to mortality in COVID-19 patients, are caused by excessive cytokines [12]. As a result, inflammatory indicators might theoretically be utilized to measure the severity of COVID-19 disease as well as the mortality risk [13].

PLR is a new inflammatory diagnostic marker that is both affordable and easily accessible in clinical settings [14]. PLR is used as a predictor of inflammation and mortality in a variety of disorders, including cardiovascular and autoimmune diseases [12, 13]. Severely ill COVID-19 patients have been reported to have high PLR levels on admission due to the quick involvement of inflammatory process-

es in COVID-19 [15, 16]. This shows that this inflammatory measure might be used to predict COVID-19 patient prognosis, especially in resource-constrained situations [8, 9].

COVID-19 disease affects bone marrow cells through CD13 receptors present on the surface of hematopoietic cells and causes hematopoietic dysfunction and decreased production. Among the hematological changes, platelets play a vital role in the disease pathogenesis, the possible mechanisms explained are as follows [2]:

1. Reduced primary platelet production due to cytokine storm and direct infection of hematopoietic precursor cells. Increased platelet destruction due to the formation of auto-antibodies. Immune complexes against platelet progenitors could also contribute to this process.

2. Increased platelet consumption by micro-thrombi formation in pulmonary capillary bed and small blood vessels.



One of the main pathogenic mechanisms explained in patients with severe COVID-19 is the activation of coagulation pathway and the formation of micro-thrombi in the lungs and blood vessels [6]. Changes in platelet parameters such as MPV and PDW indicate the activation of coagulation pathway. Large platelets in peripheral smear also reflect higher secretory activity of platelets, which in turn contributes to the hypercoagulable state [2].

Common morphological findings described among COVID-19 patients are plasmacytoid lymphocytes, large or giant platelets, and disintegrating neutrophils. A possible reason for the presence of large platelets could be the release of immature platelets from the bone marrow due to stress-induced response observed in many infections and inflammatory conditions [17]. In this study, in addition to platelet phagocytosis by activated lymphocytes, large platelets were observed among the study patients.

Zhao et al. (2020) [18] in their study observed that the value of platelet count in the early stage of the disease indicated the pathophysiological changes in COVID-19 patients. In their study, decreased platelet count was associated with increased morbidity and mortality. In the present study, there was no significant difference in PLT count at the time of admission between the ICU and non-ICU groups. However, the peak PLT value in the ICU group was higher than in the non-ICU group, which is similar to the findings of the studies conducted by Ruan et al. (2020) [19] and Lippi and colleagues [6] in China. They mentioned that thrombocytosis during the disease process was associated with a poorer prognosis.

The trend of PLT values in the ICU group in the present study is consistent with that reported in another study by Qu et al. (2020) [10] in China.

Liu et al. (2020) [20] in their study observed

that thrombocytopenia at the time of admission was associated with higher mortality and acted as an independent risk factor for COVID-19 mortality. However, in the present study, thrombocytopenia was observed only in 17.9% of admitted patients, and its correlation with mortality was not assessed.

Qu et al. (2020) [10] used PLR to assess the outcome of COVID-19 disease by comparing PLR values in severely and non-severely ill patients. They concluded that patients with a PLR cut-off value greater than 126.7 should be monitored actively to prevent further deterioration. In this study, a PLR cut-off value of 210.27 was obtained to determine the disease severity with a sensitivity of 61% and specificity of 79%.

Salamanna et al. (2020) [21] in Italy discussed the potential link between platelet parameters and COVID-19 in their review article. According to Salamanna et al., although there are many studies that suggest altered platelet parameters as indicators of aggressive COVID-19 disease, large studies stratifying patients at different stages of the disease development and analyzing the association between platelet parameters are required to develop management guidelines.

Liu et al. (2020) [20] in their study analyzed the extended PLT parameters such as MPV and PDW. They reported a significant difference in MPV between the thrombocytopenia and non-thrombocytopenia groups. In contrast, the present study showed a significant difference in PDW between the two groups. PDW is one of the markers of PLT activation and indirectly indicates hypercoagulable states as proven in many studies [6]. This explains the altered PLT activation and thrombus formation in COVID-19 patients.

Fan et al. (2020) [22] and Haung et al. (2020) [23] in their study observed lymphopenia with an absolute lymphocyte count lower than  $1 \times 10^9/L$  in ICU patients at the time of admission. The present study also showed

ALC values less than  $1 \times 10^9 / L$  predominantly in ICU patients.

Many studies [24-26] have shown increased D-dimer levels in severe infection and associated mortality. In these studies, D-dimer levels have been proposed as a prognostic marker to identify severely ill patients in the early phase. The present study also showed higher D-dimer levels in ICU patients.

Wu et al. (2020) [27] observed elevated IL-6 levels at the time of admission and its association with increased risk of death. Patients with significantly higher baseline levels of IL-6 required ICU admission compared to these with lower levels [28, 29]. The present study also showed elevated levels of IL-6 in the ICU group. However, there was no significant difference in IL-6 values between the ICU and non-ICU groups.

Fei and colleagues (2021) [30] reported higher levels of ESR in COVID-19 patients. Similar observations were made in the present study with higher ESR values in the ICU group compared to the non-ICU group.

### Conclusion

In the present study, ALC, PLR, PDW, D-dimer, and ESR were significant hematological parameters that indicated the disease severity among COVID-19 patients. Low values of ALC and high values of PLR, PDW, D-dimer, and ESR were associated with severe COVID-19 disease in ICU patients. With a cut-off value of 210.27, PLR is an easily derivable parameter that could differentiate severe COVID-19 disease from its mild and moderate forms with 79% specificity.

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**Ethical permissions:** According to the Declaration of Helsinki, the submitted research complies with all ethical statements for human studies. Ethical approval was obtained from the Institutional Human Ethics Committee (IHEC), with reference ID PSG/

IHEC/2020Apr/Exp/190.

**Conflicts of interests:** No conflict of interest was declared by the authors.

**Authors' contributions:** NG contributed to the study conception, design, and supervision, material and data collection, data analysis, literature review, writing, and critical review. ET was involved in material and data collection, data analysis, and critical review. TMSR was involved in conception, design, supervision, material collection, writing, and critical review. KS was involved in conception, design, data analysis, and critical review. LG was involved in material and data collection, data analysis, and critical review.

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**Consent to participate:** Not applicable.

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