Study of Platelet Indices as a Prognostic Marker in COVID-19 Patients

Vivek Lahane a#, Sourya Acharya a*≡ and Sunil Kumar a

a Department of Medicine, Jawaharlal Nehru Medical College, Datta Meghe Institute of Medical Sciences, Deemed to be University, Wardha, India.

Authors’ contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JPRI/2021/v33i61A35464

Open Peer Review History:

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/81054

Received 25 November 2021
Accepted 27 December 2021
Published 28 December 2021

ABSTRACT

Background: SARS-CoV-2 is a novel coronavirus responsible for the dreadful COVID-19 pandemic. In more severely ill COVID-19 patients, clotting complications, such as microvascular thrombi in the arteries and veins, are also shown. Although uncommon in the entire population of COVID-19 patients, DIC is present in more than 70 percent of dying patients and is therefore a critical feature of the final events that drive the vicious cycle culminating in death. SARS-CoV-2 is known to penetrate endothelial cells, and platelet recruitment to the sites of infection may be caused by the resulting endothelial injury. In the course of the disease, the subsequent activation and degranulation of platelets can exacerbate conditions. Prognosis of these patients is directly related to worsening of certain hematological indices. Total Platelet Count and other dynamic platelet related parameters in COVID-19 patients are currently at concern. This study aims to assess the platelet indices as a prognostic markers in COVID-19 infection.

Materials and Methods: This Cross sectional study will be conducted in COVID-19 centre of AVBRH Wardha. Total 150 confirmed COVID-19 patients will be selected, and divided into two groups based on oxygen saturation. Blood samples will be collected. They will be analysed for complete blood count, platelet indices, C-reactive protein (CRP) and for other biochemical tests. Patients with complains of shortness of breath will also be evaluated for lung computed tomography (CT). Chi square test for continuous variable and Students paired t test for numerical

# Resident;
≡ Professor;
*Corresponding author: E-mail: souryaacharya74@gmail.com;
variable will be applied. Multiple regression analysis will be applied wherever needed. SPSS 24.0 version will be used in analysis where p< 0.05 will be considered significant.

**Results:** We expect to find a significant correlation between mortality and different platelet indices.

**Conclusion:** Platelets play a vital role in prognosis of COVID-19 patients. Surveillance of platelet indices have a great prognostic value in patients with COVID-19.

**Keywords:** Platelet indices; COVID-19; SARS-COV-2; prognostic; infection; disseminated intravascular coagulation.

### 1. INTRODUCTION

A novel coronavirus, enveloped RNA betacoronavirus2, SARS-CoV-2 (severe acute respiratory syndrome coronavirus 2) emerged in Hubei region of China in December 2019[1]. It caused exponential outbreak causing pneumonia in the city of Wuhan [1]. In more severely ill COVID-19 patients, clotting complications, such as microvascular thrombi, or clots in the arteries and veins, are also shown. Another similar disease is DIC, disseminated intravascular coagulation, in which the smaller blood vessels form clots throughout the body. Although uncommon in the entire population of COVID-19 patients, DIC is present in more than 70 percent of dying patients and is therefore a critical feature of the final events that drive the vicious cycle culminating in death. A Few viruses interact with platelets and their precursor cells, megakaryocytes, leading to increased type I interferon gene expression, platelet-mediated transport, and activation of proteases. SARS-CoV-2 is known to penetrate endothelial cells, and platelet recruitment to the sites of infection may be caused by the resulting endothelial injury. In the course of the disease, the subsequent activation and degranulation of platelets can exacerbate conditions [2,3].

The infected tissue by this virus secretes molecules that activate a hyperinflammatory response, with extremely harsh amounts of cytokines and cell chemicals, in addition to the direct harm triggered by the virus, resulting in more toxicity of separate organs. This is called the cytokine storm because it is used in some other circumstances, such as cytokine release syndrome (CRS) seen in some tumours with CAR T-cell therapy. It also provides carcinogenic reactions and leads to multiorgan damage [2]. In the lung, megakaryocytes have a different presence of immune molecules, an expression other than that of the bone marrow. This may be the result of these lung cells becoming contaminated, and the result may be the release of platelets carrying the virus in the lungs. In turn, this can modify the transcription pattern to increase the development of inflammatory cytokines, also resulting in significant lung tissue injury [4].

The morbidity and mortality of COVID-19 is mostly correlated with the elderly and with preexisting comorbidities, leading to worse outcome. Despite such high morbidity and mortality, the mechanism responsible for it is currently unknown. COVID-19 patients have varied features ranging from upper respiratory tract infection associated with fever (82%) and cough (81%) to severe acute respiratory distress syndrome and sepsis. Despite this, varying lab values are found which affect patient prognosis, therapy and follow up [3].

Deranged lab values obtained in these patients are : low platelet count, low lymphocyte count and percentage, high leukocyte, high neutrophil values and ratio, reduced total protein, high D-dimer values, high C reactive protein values, higher creatinine, higher creatine kinase activity, and prolonged prothrombin time. These parameters, though found normal in mild cases during admission, worsen with the course of disease and are associated with three fourths of patients in severe condition. Prognosis of these patients is directly related to worsening of these indices [5].

Different platelet indices(PI) include: plateletcrit, mean platelet volume(MPV) and platelet distribution width(PDW). These indices are derived as a part of automatic complete blood count. There has been evidence regarding diagnostic and prognostic value of PIs in certain diseases.

Mean platelet volume(MPV) measures thrombocyte volume in femtoliters(IL); Plateletcrit (PCT) suggests volume occupied by platelets in blood, given in percentage(%) [6] given as :

\[
PCT = \frac{Platelet \ count \times MPV}{10,000} \; \quad Normal \ value: \ 0.22-0.24\%
\]
Factors such as race, age, alcohol, smoking and physical activity are responsible for modification of platelet count and MPV.

Platelet volume distribution (PDW) indicates volume variability in platelet size in percentage (%), it is found raised in presence of platelet anisocytosis. It reflects heterogeneity in platelet morphology. Reference interval of PDW reported ranges from 8.3 to 56.6%.

A marker of platelet activity, Platelet larger cell ratio (P-LCR) which is given by percentage of all platelets with a volume over 12 fL circulating in blood. Normal range is given between 15-35%. Baig studied that P-LCR is directly related to PDW and MPV and inversely related to platelet count in thrombocytopenic patients. P-LCR is observed to be more sensitive to alterations in platelet size as compared to MPV. In healthy individuals, PDW is proportionally related to MPV; however in conditions like threatened preterm labour- PDW increases and MPV decreases. Patients in vaso occlusive crises with sickle cell disease have higher PDW due to megakaryocyte hyperplasia. PCT is a biomarker of active Crohn’s disease with low high sensitivity C-reactive protein (hs-CRP).

Raised MPV and PDW are found to be associated with sepsis. In severe sepsis, PDW is said to be a poor prognostic factor. Similarly high MPV is associated with poor prognosis in pancreatic ductal adenocarcinoma and myocardial infarction. Low-grade inflammation, such as rheumatoid arthritis, can also be correlated with higher MPV levels [4,6].

The number of PLTs and dynamic parameter-related changes are therefore currently at issue in COVID-19 patients. A potential link has been established between PLT and COVID 19. severe patients. Platelet volume distribution (PDW) indicates volume variability in platelet size in percentage (%), it is found raised in presence of platelet anisocytosis. In platelet morphology, it expresses heterogeneity. The reported PDW reference interval ranges from 8.3% to 56.6%. MPV and PDW under physiological conditions are directly related. But there is contradictory evidence that indicates different underlying mechanisms with respect to the relationship between platelet volume and numbers [6].

MPV and PDW are directly related under physiological conditions. But there are conflicting evidence regarding platelet volume and platelet numbers and their relationship which signifies different underlying mechanisms. Changes in platelet activation is given by Mean platelet component (MPC) which is a measure of mean refractive index of platelets. Newer platelet activation parameters include Platelet component distribution width (PCDW) and Mean platelet mass (MPM) which are measured by Siemens Advia 120 analyser [6].

Some studies have indicated negative relation between MPV and platelet count (PC). But there is no evidence regarding MPV/platelet count ratio (MPR) and COVID count ratio (PC) [7].

Platelet indices have been previously studied in many other diseases. Platelet indices increase thrombocytopenia, type 2 diabetes mellitus and decrease in appendicitis. The overall objective of this study is to study index platelet count statistics during COVID-19 [6].

1.1 Rationale

Platelets are also shown to develop more inflammatory molecules in COVID-19 patients when activated by low levels of thrombin, a recognised source of inflammation, relative to healthy individuals who responded equally only at higher doses. Thus in COVID-19 patients, platelets tend to be primed to generate those cytokines and to release their cytokine material. COVID-19 also displays platelet degranulation, triggering an allergic response with several mediators. In both inflammatory signalling and infectious response, platelets play an important role. By integrating thrombotic and immune recruitment roles, platelets may help target haemostasis and immune responses to possible infectious agents to prevent microbial invasion. Via a variety of receptors, including Toll-like receptors, platelets directly interact with viruses. While platelets are capable of engulfing and aggregating pathogens, As demonstrated by enhanced phosphorylation of protein kinase C δ on platelets, platelet hyperactivation happens in COVID-19. This enzyme is essential to the removal of granules from platelets, activation and accumulation of platelets, and is much higher in serious patients than in non-severe cases. Platelets are shown to be primed in such a way as to display an exaggerated response to lower levels of thrombin to produce a larger number of adherent platelets, as well as to produce more inflammatory cytokines. This study is designed to look at the different effects of the covid-19 virus on the platelet system and to demonstrate with the statistical index.
1.2 Objectives

- To link platelet indices with other recognized biomarkers such as LDH, D-Dimer and the profile of coagulation.
- To compare platelet indexes with the outcomes of COVID-19 infected patients.
- To correlate the platelet indices with outcome of in terms of recovery, O2 requirement, severity and death.

2. METHODS

The research will be performed at the Hospital of Acharya Vinoba Bhave, Sawangi (Meghe). The period of the analysis will be from 2020 to 2022.

2.1 Study Design

Cross sectional observational study.

A population of COVID-19 diagnosed patients will be selected, and divided into two groups based on oxygen saturation. Blood samples will be collected. They will be analysed for complete blood count, platelet indices, C-reactive protein (CRP) and for other biochemical tests. Patients with complains of shortness of breath will also be evaluated for lung computed tomography (CT).

2.2 Setting

The thesis will be performed at the Datta Meghe Institute of Medical Sciences, J.N. Medical College, Sawangi, Wardha; a rural tertiary medical institute in central India. In Wardha district. The hospital is being planned as a care facility for COVID-19. The design for the COVID pandemic was carried out by the hospital and observation time is from June 2020. The research is observed in under the Department of Health Ministry.

2.3 Period

The research duration will be 1.5 years.

2.4 Participants

2.4.1 Inclusion criteria

Patients with covid-19 infection with positive nasopharyngeal and oropharyngeal swab samples.

2.4.2 Exclusion criteria

Hypersplenia, liver disease or cirrhosis, Other Diseases (diagnostic) Leptospirosis, Bone Marrow Disease.

Bias: No bias.

2.5 Study Size

The surveys performed to measure a population parameter are cross sectional, such as the prevalence of some disease in a group or the average value of some quantitative attribute in a population. The sample size formula is different for the qualitative variable and the variable quantity.

2.6 Statistical Methods

Test for statistical analysis: Chi square test for continuous variable and Students paired t test for numerical variable. Multiple regression analysis will be applied wherever needed. SPSS 24.0 version will be used in analysis where p< 0.05 will be considered significant.

3. EXPECTED OUTCOMES/RESULTS

A variety of proportional hazard regression models were adapted in this study for several risk factors associated with death at admission, including age, sex, baseline, comorbidity, lymphocyte count, blood lactate, c-reactive protein, glucocorticoid therapy, and intravenous immunoglobins.

For mortality, the result suggested that. The number of platelets has been dramatically related to a 40 percent decrease in mortality, but the number of platelets raises the chance of death during therapy.

4. DISCUSSION

In a study done by Ertugrul Guclu et.al in COVID-19 patients, it was derived that oxygen saturation during admission and MPV difference between first and third day of hospitalization were significant parameters for predicting mortality [8]. In patients with oxygen saturation under 90% during hospital admission, mortality rates were 8.4 times higher whereas 1 unit increase in MPV lead to 1.76 times increased mortality. Thus it was concluded that with lung capacity of patients an additional parameter of Mean platelet volume should be considered to predict mortality in COVID-19 patients [9].

In a study done by Yaxin Wang et al it was concluded that in patients with COVID-19 thrombocytopenia is commonly associated with
increased risk of mortality. It was observed that with decrease in platelet count the mortality increased [9]. It has been also found that pulmonary capillary autopsies contain megakaryocytes which are known to be associated with innate immunity functions [10]. The overall analysis of patients showed that high platelet turnover is consistent with macro thrombocyte production. Dense granules, and platelet aggregate found in patients show increased platelet activity [11]. In ICU, thrombocytopenia occurs due to endogenous and iatrogenic factors. Surprisingly, it has been observed that thrombocytopenia is uncommon in severe COVID-19 infections. Blood films of COVID positive ICU patients and of COVID-19 non positives were examined for platelet aggregates and macro thrombocytes which showed increased platelet activity. These results were compared with non severe COVID-19 blood films which didn’t show similar findings [11]. Few of the related studies were reviewed [12-19]. This study has been designed to understand and correlate outcome of patients infected with SARS-COV-2 [20-27].

5. CONCLUSION

COVID-19 has emerged to be a pandemic which started from city of Wuhan, china and created an outbreak worldwide. Its symptoms have been ranged from mild to severe respiratory symptoms ranging from cough and fever to severe acute respiratory distress leading to increased intensive care unit admissions ultimately leading to high morbidity and mortality. These rates were found higher in patients with associated or pre existing medical conditions. Recent studies have tried to correlate between mortality in COVID-19 to various hematological indices. It has been proved that platelets develop more inflammatory molecules in these patients when activated by low thrombin levels which causes inflammation, as compared to healthy individuals who responded equally only on higher doses. Thus, platelets play a vital role in prognosis of COVID-19 patients. Hence this study has been designed to study different effects of COVID-19 on platelet system and to correlate various platelet indices, statistically with outcome of these patients.

6. LIMITATIONS

The main point we can claim is financial constraints, but the number of patients for the research is also a problem throughout the study, the perfect criteria require a perfect observation sample, the MPV and PDW are the two parameters we used that differentiate between COVID patients cost-effectively, but easily and explicitly, to illustrate the greater seriousness of this disease, we required the large number of patients.

GENERALIZABILITY

This study has been conducted over a small population. Hence it cannot be generalized to the community.

CONSENT

As per international standard or university standard, patients’ written consent will be collected and preserved by the author(s).

ETHICAL APPROVAL

As per international standard or university standard written ethical approval will be collected and preserved by the author(s).

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Coronavirus Disease (COVID-19) Situation Reports [Internet]. [cited 2020 Dec 7]. Available:https://www.who.int/emergencies/diseases/novel-coronavirus-2019/situation-reports
2. Drosten C, Günther S, Preiser W, van der Werf S, Brodt H-R, Becker S, et al. Identification of a novel coronavirus in patients with severe acute respiratory syndrome. N Engl J Med. 2003; 348(20):1967–76.
3. Velavan TP, Meyer CG. The COVID-19 epidemic. Trop Med Int Health. 2020 Mar;25(3):278–80.
4. Chan AS, Rout A. Use of Neutrophil-to-Lymphocyte and Platelet-to-Lymphocyte Ratios in COVID-19. J Clin Med Res. 2020;12(7):448–53.
5. Lopez E, Bermejo N, Berna-Enro A, Alonso N, Salido GM, Redondo PC, et al. Relationship between calcium mobilization and platelet α- and δ-granule secretion. A role for TRPC6 in thrombin-evoked δ-
granule exocytosis. Arch Biochem Biophys. 2015;585:75–81.

6. WHO. Pneumonia of unknown cause – China [Internet]. WHO. World Health Organization; [cited 2020 Dec 7]. Available: http://www.who.int/csr/don/05-january-2020-pneumonia-of-unknown-cause-china/en/

7. Nugent D, McMillan R, Nichol JL, Slichter SJ. Pathogenesis of chronic immune thrombocytopenia: increased platelet destruction and/or decreased platelet production. Br J Haematol. 2009;146(6):585–96.

8. Platelet Concentrates in Musculoskeletal Medicine [Internet]. [cited 2020 Dec 7]. Available: https://pubmed.ncbi.nlm.nih.gov/15094938/

9. Warkentin TE, Aird WC, Rand JH. Platelet-endothelial interactions: sepsis, HIT, and antiphospholipid syndrome. Hematol Am Soc Hematol Educ Program. 2003;497–519.

10. Plateletcrit, mean platelet volume, platelet distribution width: its expected values and correlation with parallel red blood cell parameters - PubMed [Internet]. [cited 2020 Dec 8]. Available: https://pubmed.ncbi.nlm.nih.gov/15094938/

11. Regulation of Chronic Wounds with Allogeneic Platelet Gel Versus Hydrogel Treatment: A Prospective Study [Internet]. [cited 2020 Dec 7]. Available: https://pubmed.ncbi.nlm.nih.gov/15094938/

12. Gupta AR, Sarode, S. Kumar, and G.M. Dhopavkar. “Impact of Platelet Indices as Prognostic Markers of Sepsis.” International Journal of Pharmaceutical Research. 2019;11(3):1413–17. Available: https://doi.org/10.31838/ijpr/2019.11.03.153.

13. Walunjkar, R.S., S. Khadge, S. Kumar, S. Bawankule, and S. Acharya. “Platelet Indices as a Predictor of Microvascular Complications in Type 2 Diabetes.” Indian Journal of Endocrinology and Metabolism. 2019;23(2):206–10. Available: https://doi.org/10.4103/ijem.IJEM-13-19.

14. Khatib MN, Gaidhane S, Khatib M, Ahmed M, Gaidhane A, Syed ZQ. SARS-CoV and SARS-CoV-2: Similar Viruses with Different Trajectories. Wutan Huatan Jisuan Jishu. 2010;16(5):544–48.

15. Jaiswal A, Borage S, Shelotkar P. A Clinical Approach to COVID-19. International Journal of Research in Pharmaceutical Sciences. 2020;11(1):723–29. Available: https://doi.org/10.26452/ijrps.v11i1SPL1.3073.

16. Khan SS, Quazi, Kaple M. The Demographical and Epidemiological Profile of Coronavirus Disease 2019 (Covid-19): A Review. Journal of Critical Reviews. 2020;7(10):4–8. Available: https://doi.org/10.31838/jcr.07.10.02.

17. Padole VS, Kalsait RP, Ambad R, Kute P. Effect of COVID 19 Affecting Geriatric Patients. International Journal of Current Research and Review. 2020;12(17):182–87. Available: https://doi.org/10.31782/IJCRRR.2020.121729.

18. Dhar R, Singh S, Talwar D, Mohan M, Tripathi SK, Swarnakar R, Trivedi S, Rajagopala S, D'Souza G, Padmanabhan A, Baburao A. Bronchiectasis in India: results from the European multicentre bronchiectasis audit and research collaboration (EMBARC) and respiratory research network of India registry. The Lancet Global Health. 2019;7(9):e1269-79.

19. Prasad N, Bhatt M, Agarwal SK, Kohli HS, Gopalakrishnan N, Fernando E, Sahay M, Rajapurkar M, Chowdhary AR, Rathi M, Jeloka T. The adverse effect of COVID pandemic on the care of patients with kidney diseases in India. Kidney International Reports. 2020;5(9):1545-50.

20. Walia IS, Borle RM, Mehendiratta D, Yadav AO. Microbiology and antibiotic sensitivity of head and neck space infections of odontogenic origin. Journal of Maxillofacial and Oral Surgery. 2014;13(1):16-21.

21. Lohe VK, Degwekar SS, Bhowate RR, Kadu RP, Dangore SB. Evaluation of correlation of serum lipid profile in patients with oral cancer and precancer and its association with tobacco abuse. Journal of Oral Pathology & Medicine. 2010;39(2):141-8.

22. Korde S, Sridharan G, Gadbail A, Poornima V. Nitric oxide and oral cancer: A review. Oral oncology. 2012;48(6):475-83.

23. Gondivkar SM, Gadbail AR. Gorham-Stout syndrome: a rare clinical entity and review of literature. Oral Surgery, Oral Medicine,
24. Gadbail AR, Chaudhary M, Gawande M, Hande A, Sarode S, Tekade SA, Korde S, Zade P, Bhowate R, Borle R, Patil S. Oral squamous cell carcinoma in the background of oral submucous fibrosis is a distinct clinicopathological entity with better prognosis. Journal of Oral Pathology & Medicine. 2017;46(6):448-53.

25. Gadre PK, Ramanojam S, Patankar A, Gadre KS. Nonvascularized bone grafting for mandibular reconstruction: myth or reality?. Journal of Craniofacial Surgery. 2011;22(5):1727-35.

26. Sorte K, Sune P, Bhake A, Shivkumar VB, Gangane N, Basak A. Quantitative assessment of DNA damage directly in lens epithelial cells from senile cataract patients. Molecular Vision. 2011;17:1.

27. Basak S, Rajurkar MN, Mallick SK. Detection of Blastocystis hominis: a controversial human pathogen. Parasitology Research. 2014;113(1):261-5.