

Neutrophil-to-lymphocyte ratio, platelet-to-lymphocyte ratio, and absolute lymphocyte count as mortality predictor of patients with Coronavirus Disease 2019

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ABSTRACT

Introduction: Coronavirus Disease 2019 (COVID-19) has a various spectrum of symptoms from asymptomatic to severe. In previous research, hematological examination could predict the severity of the infection. This study is aimed to evaluate Neutrophil-to-Lymphocyte Ratio (NLR), Platelet-to-Lymphocyte Ratio (PLR), and Absolute Lymphocyte Count (ALC) as a predictor of COVID-19 mortality.

Materials and Methods: The study was a cross sectional research with retrospective design by collecting data from medical records of the COVID-19 patients at a General Hospital Anwar Medika of East Java, for the period June 2020 to January 2021. A comparative test was performed using the Mann-Whitney test. The predictive ability was assessed using the ROC curve.

Results: A total of 70 subjects were involved in this study with 16 (22.9%) mortality. And the average NLR and PLR were 4.11 and 174.39. The Mann-Whitney test showed significant results ($p < 0.05$) on leukocytes, NLR, PLR, and ALC of the patients. The ROC curve showed that under the curve of leukocytes, NLR, PLR, and ALC were 0.749, 0.731, 0.719, and 0.306, respectively.

Conclusion: With higher sensitivity and specificity by leukocytes 75% and 70.4%. Leukocytes, NLR, and PLR showed good ability to predict patient mortality from COVID-19.

KEYWORDS:

Coronavirus, mortality, predictive tools

INTRODUCTION

Coronavirus is a family of viruses found in humans and animals, the emergence of a new virus outbreak belonging to the coronavirus group was first discovered in the city of Wuhan, China. Within a few weeks, this virus spread to various parts of the continent. On February 20, 2020, the World Health Organization (WHO) announced that the outbreak caused by this new type of Coronavirus, namely Coronavirus Disease 2019 (COVID-19), was global. It is recorded that at this time, around 184 million people have

been affected, and almost 4 million deaths due to Coronavirus, which has infected 222 countries around the world.¹

In Indonesia alone, there were 2.3 million confirmed cases with nearly 60,000 deaths.² In SARS-CoV-2 infection, there are changes in several laboratory indicators which are also found in several other viral infections, because the hematopoietic system is a system that feels the impact of a viral infection.^{3,4} Several previous studies have shown that there is lymphocytopenia, neutrophilia, eosinopenia, thrombocytopenia, or thrombocytosis, as well as the presence of reactive lymphocytes which indicate an acute infection.⁴ Apart from the changes in the results of the hematological examination, the results can also be used to predict the severity of COVID-19. A meta-analysis stated that leukocytosis, lymphopenia, and thrombocytopenia are predictors of the severity of COVID-19 and increase the incidence of mortality with these results.⁵

Several other studies have also shown that NLR, PLR, and ALC can predict mortality from COVID-19 infection.^{6,7} In this study, we evaluated the COVID-19 mortality hematologic predictors at Anwar Medika General Hospital as one of the COVID-19 referral hospitals in Indonesia

MATERIAL AND METHODS

The study used an observational analytic cross-sectional research with retrospective design. The profile description is carried out retrospectively by collecting data from medical records of the COVID-19 patients' data consisting of age, gender, leukocytes and the differential count, platelets, length of stay, and mortality status. The hematological result was then calculated to produce NLR (neutrophil-to-lymphocyte ratio), PLR (platelet-to-lymphocyte ratio), and ALC (absolute lymphocyte count).

These were the following equation:

$$NLR = \frac{NLR = (\text{Absolute neutrophil count (Cells}/\mu\text{L)})}{(\text{Absolute lymphocyte count (Cells}/\mu\text{L)})}$$

$$PLR = \frac{\text{Thrombocyte count (Cells}/\mu\text{L)}}{\text{Absolute lymphocyte count (Cells}/\mu\text{L)}}$$

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The sample used in this study were patients infected with COVID-19 at the Anwar Medika General Hospital during the period of June 2020 to January 2021. With the inclusion criteria of patients who have been confirmed positive for COVID-19 through PCR examination results, while patients with a history of blood transfusions were excluded from this study.

Furthermore, comparative tests were conducted between the hematological profile to the length of stay (LoS) and patient mortality. Comparative tests were conducted using a two-sample independent t2 test on data with a normal distribution and using the Mann-Whitney test on data with an abnormal distribution. Variables that have significant values were then tested for predictive ability using the ROC curve then interpreted as sensitivity and specificity. The highest sensitivity and specificity are obtained by the curve reading that produced by IBM® SPSS®

RESULTS

A total of 70 subjects were involved in this study. From the subjects, 10 subjects (14.3%) were males, and the rest were females (85.7%). The average age of the research subjects was 51.03 years with the youngest being 30 years old and the oldest 82 years old.

From the results of the hematological examination, the average leukocytes, platelets, neutrophils, and lymphocytes were 9,730/ μ L, 265,671/ μ L, 5,960/ μ L, and 1,830/ μ L. And from the calculation, the average NLR and PLR were 4.11 and 174.39. With an average length of stay (LoS) of 10.91 days.

From the mortality data, 16 patients (22.9%) died. The average age of the subjects who died was 53.19 years, higher than the group who survived 50.58 years. From the calculation of leukocytes, platelets, neutrophils, NLR, and PLR, it was found that the average death group was higher than the survival group. Meanwhile, ALC and LoS were lower in the mortal group than in the surviving group. The comparative test showed significant results ($p < 0.05$) on leukocytes, NLR, PLR, ALC, and length of stay (LoS) of the patients.

The results of the receiver operating characteristic (ROC) curve the largest area under the curve (AUC) was produced by leukocytes, 0.749, followed by NLR and PLR. Meanwhile, ALC has the lowest AUC of 0.306. From the results of the highest sensitivity obtained PLR of 81.3% and the highest specificity is 70.4% leukocytes.

DISCUSSION

The number of deaths due to COVID-19 is increasing every day in Indonesia, with the number of deaths reaching a thousand on 07/08/2021.² In addition, there are also more than 300,000 active cases of confirmed patients infected with COVID-19, which further reduces the ability of hospitals to handle patients.² However, clinical manifestations of the confirmed patients vary from asymptomatic to severe symptoms.^{8,9} Because of this, it is necessary to stratify individuals with confirmed COVID-19 to optimize the capacity of the available hospitals.

From our research, it was found that from 70 subjects consisted of 16 subjects (22.9%) who died during the observation and the rest recovered. The mortality rate of 22.9% was higher if were looked at the death rate throughout Indonesia, which was 2.6%.² This finding could be related to community stigma regarding COVID-19 which causes patients to be late in seeking help to health centers which is associated with increased patient mortality.^{10,11}

From the findings in our study, it was found that the average age of the covid patients we treated was 51.03 years, which is the age group at risk of severe infection in COVID-19.¹²⁻¹⁴ This result is also supported by the finding of an older age in mortality cases compared to the survivors. However, in our study, there were no significant results between age and mortality status which was not in line with previous studies which stated that age was associated with patient mortality.¹⁴

The comparison test result from mortality status and hematological results were found that there was a relationship between mortality status with leukocytes, NLR, PLR, and ALC. There was also a significant relationship between mortality status and length of stay (LoS) of the patients. In the previous study, evaluation of the hematological component compared to the severity of COVID-19 infection showed that there was a relationship between either NLR, PLR, or ALC and infection severity with various degrees of association.¹⁵⁻¹⁷ Our findings showed that leukocytes, NLR, and PLR were higher in subjects with mortality compared to surviving subjects. While the ALC showed the opposite results which decreased in subjects who experienced mortality. This finding is in line with research.¹⁸ So that leukocytes, NLR, PLR, and ALC could be used as predictors of severity and mortality from COVID-19 patients. From the ROC curve, it was found that leukocytes, NLR, and PLR had AUC more than 0.50, but not with ALC. From the results of the best sensitivity and specificity, leukocytes were obtained with a sensitivity of 75% and a specificity of 70.4%, followed by PLR with a sensitivity of 81.3% and a specificity of 63%. Although ALC has a relationship with the patient's mortality status, the predictive ability of ALC on mortality status is quite low, seeing the AUC of only 0.306. In the previous predictive value testing in the research conducted by Yang et al. the sensitivity from NLR and PLR were 88% and 77%, respectively.⁶ These results indicate that this modality can be used as a basis for predicting mortality in COVID-19 patients.

The association of leukocytes and NLR in infection cases in previous studies has been associated with the occurrence of cytokine storms that often occur in severe COVID-19 patients.¹⁹ Although in this study no further investigation was carried out regarding the cause of death suffered by the patients, the increase in the number of leukocytes, which were dominated by polymorphonuclear cells, indicated that this finding was a predictor of increased immune activity in the patients' body which could be a sign of a cytokine storm. In line with the investigation Xiong et al. stated that the increase in neutrophil was a result of the release of CXCL-2 and CXCL-8 which increased neutrophil recruitment and indicated high cytokine activity in the patient's body.²⁰

From our findings, it was found that there was a higher PLR

Table I: Characteristic of the subject

Variable	Mortality status	
	Survive, n (%)	Mortal, n (%)
Gender		
Male	7 (70%)	3 (30%)
Female	47 (78.3%)	13 (21.7%)

Table II: Comparison variable between mortality status

Variable	Mean±SD			p
	Total subject	Survived subject	Dead subject	
Age (years old)	51.03±11.77	50.58±12.814	53.19±7.332	NS
Leukocyte (μL)	9,730±4,720	8,672±3,687	13,200±6,219	0.003
Thrombocyte (μL)	265,671±101,263	259,113±106,998	286,812.5±82,743.1	NS
Neutrophil (μL)	5,960±3,310	5,183.8±2,287.4	8,505.3±4,820.2	NS
NLR	4.11±3.40	3.23±2.10	11±5.02	0.005
PLR	174.40±97.80	160.76±94.797	223.55±96.64	0.008
ALC (μL)	1,830±981.5	1,895.4±967.3	1,572.4±1,035.8	0.019
Length of stay (days)	10.91±4.52	11.91±4.42	75±3.45	0.001

NS, Not significant

Table III: The highest sensitivity and specificity of the variable

Predictive variable	AUC	Sensitivity	Specificity
Leukocyte	0.749	75%	70.4%
NLR	0.731	75%	57.4%
PLR	0.719	81.3%	63%
ALC	0.306	56.3%	20.4%

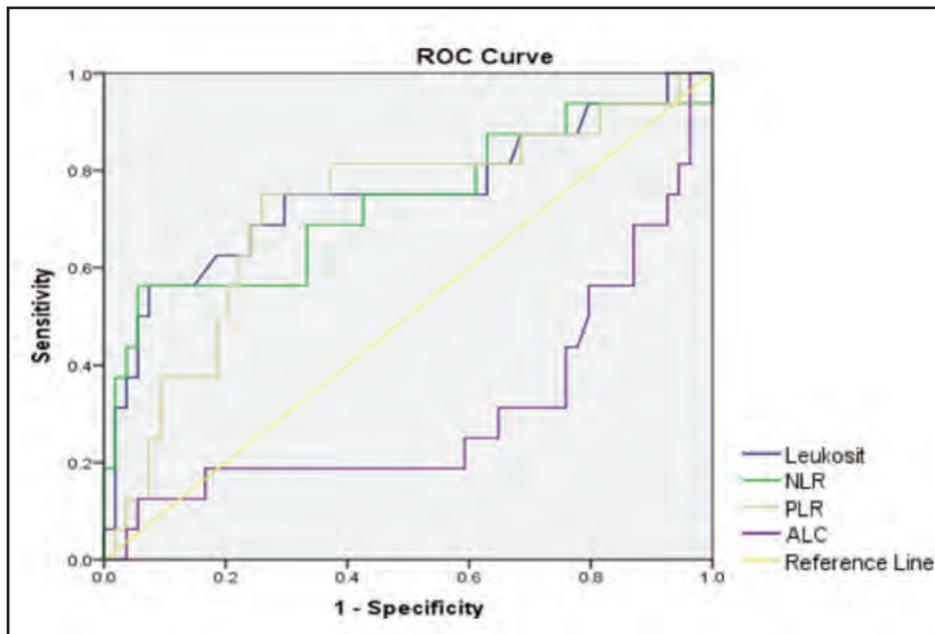


Fig. 1: ROC Curve of the predicting variable

in the mortal group compared to the surviving group. This finding is in line with previous research.²¹ However, the increase in PLR was not in line with increase in platelets, there was no significant difference between the survivors and the mortality group from the platelet count. The increase in PLR is due to a decrease in the denominator component which is ALC. Several hypotheses that the decrease in ALC in COVID-19 patients is due to the P53 signaling process and FAS signaling which causes lymphocytes to undergo apoptosis.^{20,22}

The findings in this study can be used to detect the possible severity and mortality of COVID-19 patients. So that it can be used as consideration for determining the room and therapy to be given to patients. However, this study has limitations mainly because the focus of this study was to evaluate the predictive ability of NLR, PLR, and ALC on COVID-19 mortality in one of the COVID-19 referral sites in Indonesia. Therefore, the subjects involved in this study are limited and cannot represent subjects in Indonesia in general. However, this study can be the basis that NLR, PLR, and leukocytes can be used as predictors of COVID-19 mortality that are easy and inexpensive.

CONCLUSION

Hematological components (Leukocytes, NLR, PLR, and ALC) have a relationship with mortality status in COVID-19 patients. However, only leukocytes, NLR, and PLR showed good ability to predict patient mortality so it could be used as a triage consideration for COVID-19 patients in the conclusions with the goals of the study but avoid unqualified statements and conclusions not completely supported by the data.

REFERENCES

- American Library Association. Covid-19 Coronavirus Pandemic [cited 2020] Accessed from: <https://www.worldometers.info/coronavirus/>
- GugusTugas COVID-19. Data SebaranKasusPositif [cited2020] Accessed from: <https://covid19.go.id/>
- Debut B,Smadja DM. Is COVID-19 a new hematologic disease? *Stem Cell Rev Reports* 2021;17(1):4–8.
- de Oliveira Toledo SL, Nogueira LS, das Graças Carvalho M, Rios DRA, de Barros Pinheiro M. COVID-19: Review and hematologic impact. *Clin Chim Acta* 2020;510:170–6.
- Henry BM, De Oliveira MHS, Benoit S, Plebani M, Lippi G. Hematologic, biochemical and immune biomarker abnormalities associated with severe illness and mortality in coronavirus disease 2019 (COVID-19): A meta-analysis. *Clin Chem Lab* 2020; 58: 1021–8.
- Yang AP, Ping LJ, Qiang TW, Ming LH. The diagnostic and predictive role of NLR, d- NLR and PLR in COVID-19 patients. *Int Immunopharmacol* 2020;84.
- Wagner J, DuPont A, Larson S, Cash B,FarooqA. Absolute lymphocyte count is a prognostic marker in Covid-19: A retrospective cohort review. *Int J Lab Hematol* 2020;42(6):761–5.
- Hafez MAF. The mean severity score and its correlation with common computed tomography chest manifestations in Egyptian patients with COVID-2019 pneumonia. *Egypt J RadiolNucl Med* 2020;51(1).
- Storch-de-Gracia P, Leoz-Gordillo I, Andina D, Flores P, Villalobos E, Escalada-Pellitero S, et al. Clinical spectrum and risk factors for complicated disease course in children admitted with SARS-CoV-2 infection. *An Pediatr* 2020;93(5):323–33.
- Abdelhafiz AS,Alorabi M. Social stigma: The hidden threat of COVID-19. *Front in Public Health* 2020; 8.
- Alaa A, Qian Z, Rashbass J, Bengier J, van der Schaar M. Retrospective cohort study of admission timing and mortality following COVID-19 infection in England. *BMJ Open* 2020;10(11):e042712.
- Wolff D, Nee S, Hickey NS, Marschollek M. Risk factors for Covid-19 severity and fatality:a structured literature review. *Infection* 2021; 49: 15–28.
- Li X, Xu S, Yu M, Wang K, Tao Y, Zhou Y, et al. Risk factors for severity and mortality in adult COVID-19 inpatients in Wuhan. *J Allergy Clin Immunol* 2020;146(1):110–8.
- Zheng Z, Peng F, Xu B, Zhao J, Liu H, Peng J, et al. Risk factors of critical & mortal COVID- 19 cases: A systematic literature review and meta-analysis. *J Infect* 2020; 81: e16–25.
- Imran MM, Ahmad U, Usman U, Ali M, Shaukat A,Gul N.Neutrophil/lymphocyte ratio—A marker of COVID-19 pneumonia severity. *Int J Clin Pract* 2021;75(4).
- Qu R, Ling Y, Zhang Y, Wei L, Chen X, Li X, et al. Platelet-to-lymphocyte ratio is associated with prognosis in patients with coronavirus disease-19. *J Med Virol* 2020;92(9):1533–41.
- Zeng F, Li L, Zeng J, Deng Y, Huang H, Chen B, et al. Can we predict the severity of coronavirus disease 2019 with a routine blood test? *Polish Arch Intern Med* 2020;130(5):400–6.
- Illg Z, Muller G, Mueller M, Nippert J, Allen B. Analysis of absolute lymphocyte count in patients with COVID-19. *Am J Emerg Med.* 2021;46:16–9.
- Wang J, Jiang M, Chen X, Montaner LJ. Cytokine storm and leukocyte changes in mild versus severe SARS-CoV-2 infection: Review of 3939 COVID-19 patients in China and emerging pathogenesis and therapy concepts. *J Leukoc Biol* 2020;108: 17–41.
- Xiong Y, Liu Y, Cao L, Wang D, Guo M, Jiang A, et al. Transcriptomic characteristics of bronchoalveolar lavage fluid and peripheral blood mononuclear cells in COVID-19 patients. *Emerg Microbes Infect* 2020;9(1):761–70.
- Lin S, Mao W, Zou Q, Lu S, Zheng S. Associations between hematological parameters and disease severity in patients with SARS- CoV-2 infection. *J Clin Lab Anal* 2021;35(1).
- Feng Z, Diao B, Wang R, Wang G, Wang C, Tan Y, et al. The novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) directly decimates human spleens and lymph nodes. *medRxiv* 2020.