The Correlation between Neutrophil-to-Lymphocyte Ratio with C-reactive Protein and D-dimer Level among Indonesian COVID-19 Cases

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Abstract

BACKGROUND: Coronavirus disease-19 (COVID-19) pandemic has resulted high number of mortalities globally. Several inflammatory and coagulation biomarkers have been studied for predicting and differentiating severe COVID-19 such as C-reactive protein (CRP) and D-dimer. However, those markers may not readily available in developing countries.

AIM: The aim of the study was to assess the utility of neutrophil-to-lymphocyte ratio (NLR), a widely available and inexpensive laboratory examination, as reliable inflammatory biomarkers for Indonesian COVID-19 patients; by analyzing the correlation of NLR level with CRP and D-dimer plasma level.

METHODS: We conducted cross-sectional study in Professor Dr. R.D. Kandou Hospital, Manado involving RT-PCR confirmed and hospitalized COVID-19 patients. Lymphocyte count, NLR, CRP, and D-dimer were examined in severe and non-severe COVID-19 cases at hospital admission. Correlation test was done using Spearman correlation test.

RESULTS: A total of 40 COVID-19 patients were included in the analysis, with 50% having mild disease and other half having severe disease. The NLR, CRP, and D-dimer were significantly higher in severe and non-severe COVID-19 cases at hospital admission. Significant correlation was found between NLR level with CRP (p = 0.001 and r = 0.506) and also with D-dimer level (p = 0.000 and r = 0.570) in differentiating severity of COVID-19.

CONCLUSION: NLR is correlated with CRP and D-dimer level; therefore, NLR may serve as reliable, cost-effective, and practical inflammatory biomarker for differentiating severe and non-severe COVID-19 cases.

Introduction

Coronavirus disease-19 (COVID-19) is an ongoing global pandemic which has resulted over 273 million infected cases and more than 5.3 million deaths according to the World Health Organization (WHO) as of December 19, 2021 [1]. The number of new and death cases is predicted to be still fluctuating due to the incoming of new COVID-19 variants, wearing of protective antibody titer, and other factors. Unfortunately, until now, there are still no specific anti-SARS-CoV-2 drugs. The most of COVID-19 death cases were attributed to multiple organ failure due to cytokine storm and pulmonary embolism [2]. Several efforts have been employed to reduce those fatally COVID-19 complications, such as the use of dexamethasone and anticoagulant prophylaxis for hospitalized COVID-19 patients [3]. The evidence of cytokine storm presence in COVID-19 was proved by many studies which shown by high inflammatory cytokines and biomarkers such as interleukin-6 (IL-6), interferon (IFN)-α, IFN-γ, C-reactive protein (CRP), and others. C-reactive protein has been shown as reliable early biomarker to predict risk for severity of COVID-19 [4], [5]. Therefore, patients with the high level of CRP level on admission may be placed for closer observation. D-dimer is widely known as marker of inflammation and presence of thrombus. In COVID-19 cases, elevated D-dimer was associated with more severe COVID-19 disease course and higher risk of mortality [6]. While CRP and D-dimer are becoming an established COVID-19 prognostic biomarkers, those biomarkers examination may not be applicable in low- and middle-income countries or in limited resources health-care facilities. A cost-effective yet readily available and accurate biomarker is needed. Therefore, we would like to assess the utility of neutrophil-to-lymphocyte ratio (NLR),
a simple, widely available, and inexpensive laboratory examination, as reliable inflammatory biomarkers for Indonesian COVID-19 patients; by comparing NLR level with CRP and D-dimer plasma level [7].

Methods

We conducted a cross-sectional study in Professor. Dr. R. D. Kandou Hospital, Manado, North Sulawesi, Indonesia and recruited confirmed mild and severe hospitalized COVID-19 patients aged older than 18 years old from June to August 2020. We excluded patients with waist circumference below 90 cm, patients with cancer, Human Immunodeficiency Virus (HIV) infection, and autoimmune disease. The disease severity was defined using National Guidelines of COVID-19 Management [8]. Mild case was defined as COVID-19 patients with the upper respiratory tract infection symptoms, without pneumonia and the need for oxygen supplementation. Severe case was defined as severe pneumonia which reflected by fever, cough, dyspnea, having respiratory rate more than 30 times/min, severe respiratory distress, and oxygen saturation below 90%. Confirmation of COVID-19 was based on Real-Time-Polymerase-Chain-Reaction (RT-PCR) testing. Informed consent was taken before the study and samples were collected with consecutive sampling methods. Sera were collected on the admission day. Each patient’s demographic, clinical, and hematological data were recorded. The NLR was calculated by dividing the number of neutrophils to lymphocyte count. The CRP value was counted using immunoturbidimetric method. Normal CRP value was defined as lower than 5 mg/dl. D-dimer level was quantified using Enzyme-linked Fluorescent Immunoassay (ELFA). D-dimer count was normal if below 0.5 µg/ml.

Data analysis was supported with Statistical Package for the Social Science (SPSS) version 22. Continuous variables were denoted as mean ± standard deviation. Normality test was conducted using Shapiro–Wilk test. Unpaired t-test or Mann–Whitney test was used to compare the mean of NLR, D-dimer, and CRP between mild and severe COVID-19 group. Pearson or Spearman correlation test was used to determine the correlation between NLR and D-dimer and CRP value.

Results

We recruited 40 COVID-19 patients during study period with half of them were having severe disease and 50% were male. The mean age of patients was 53.53 ± 15.15 years old. According to the disease severity, we found that 20 patients were having mild disease and 20 patients were having severe COVID-19 disease. The overall mean of absolute lymphocyte count was 1790.87 ± 1018.78 cells/mm³. The overall mean of NLR was 7.38 ± 6.94. The overall mean of CRP and D-dimer was 23.73 ± 19.39 mg/dl and 4.88 ± 6.36 µg/ml, respectively. Detail regarding our subject’s characteristics and comparison between mild and severe COVID-19 group can be seen in Table 1.

In our study, the Mann–Whitney test showed that NLR value was significantly higher in severe COVID-19 disease group compared to mild COVID-19 group with p value of 0.002. Similar finding was also found in analysis of CRP (p = 0.000) and D-dimer (p = 0.001). The Spearman correlation test showed significant correlation between NLR and CRP level (p = 0.001) with correlation coefficient of 0.506. The correlation analysis also showed significant correlation (p = 0.000) between NLR and D-dimer level correlation coefficient of 0.570.

Discussion

The NLR role as prognostic marker for COVID-19 patients was extensively studied. NLR may become a reliable, accessible, and cost-effective inflammation parameters, especially in developing country such Indonesia during COVID-19 pandemic era. NLR was easily calculated at emergency department using routine laboratory test [9], [10]. A meta-analysis by Simadibrata et al. which involving more than 5000 COVID-19 patients found that the higher NLR levels on admission were associated with 2.7 times higher mortality risk compared to patients with normal NLR. The higher NLR level was also associated with more severe disease COVID-19 course [7]. This finding was in line with our result which the higher NLR value is more commonly found in severe COVID-19 group. Elevation of NLR value may be explained by the increased level of neutrophil due to inflammation response and due to the lymphocytopenia. Lymphocytopenia occurred in 90% of severe COVID-19 cases [11], [12], [13]. Systematic review by Zhao et al. concluded that COVID-19 patients with lymphocytopenia are associated with nearly 3 times increased risk for severe disease course [12]. Several mechanisms have been proposed regarding the occurrence of lymphocytopenia in COVID-19 patients such as SARS-CoV-2 may induce apoptosis and pyroptosis of lymphocytes, bone marrow suppression due to proinflammatory cytokines released, thymus suppression, activation-induced cell death of lymphocytes, tissue redistribution of lymphocytes, and several other pathways [11].

In our study, we found significant correlation with moderate strength between NLR and CRP value. The CRP which determined as acute phase reactant plasma
protein and mainly produced in liver was induced by several inflammatory cytokines such as IL-6. The use of CRP as inflammation biomarker has been established. The CRP value may decrease gradually over 18–20 h after inflammatory stimuli were stopped [14]. The use of CRP in COVID-19 case management has been proposed as early marker for predicting severe COVID-19. Aji found that CRP threshold value of 26.9 mg/L may serve as optimal cutoff and every one-unit increase in CRP may lead to 5% increased risk for getting severe COVID-19 [5]. Smiłowitz et al. found that elevated CRP level was associated with worse clinical outcome of COVID-19 which defined as more critical illness, higher in-hospital mortality, venous thrombo-embolism, acute kidney injury rate [15]. We also found higher CRP value in severe COVID-19 group in our study. We also found that NLR was correlated with CRP value which associated with more severe COVID-19 at higher level plasma level. Similar finding was also found by Mousavi-Nasab et al., which NLR was positively correlated with CRP value (r=0.23) [16]. Study in Indonesia conducted by Sukrisman et al. also showed strong correlation between NLR and CRP (p < 0.001, r = 0.738) [17]. All these findings showed that NLR may serve as suitable inflammation marker in remote area for predicting severe COVID-19.

Mortality in COVID-19 cases may attribute due to venous thromboembolism events caused by hypercoagulability condition. D-dimer value is regarded as hemostasis activation and thrombosis marker. Meta-analysis by Zhan et al. showed that D-dimer may predict severe cases of COVID-19 with moderate accuracy [18]. Another systematic review by Rostami et al. also found that increased 3–4 fold of D-dimer level was linked to poor prognosis and they recommended to measure D-dimer level in COVID-19 patients [19]. Poudel et al. proposed that D-dimer cutoff of 1.5 µg/ ml on admission is the optimal cutoff for predicting mortality in COVID-19 cases [20]. In this study, we found significantly higher D-dimer level in severe COVID-19 group. Isbaniah et al. found that NLR and D-dimer value on admission was important for predicting clinical outcome of Indonesian COVID-19 patients [21]. Man et al. found that NLR was significantly correlated with D-dimer in predicting COVID-19 severity (p < 0.001 and r = 0.49) [22]. This finding is similar to our finding.

The limitation of our study is regarding the small number of subjects; therefore, further study with larger sample size is warranted to establish stronger correlation between NLR, CRP, and D-dimer level among Indonesia COVID-19 patients.

**Conclusion**

NLR was found well correlated with established inflammatory and coagulation marker which capable in predicting severe COVID-19. Therefore, NLR which easily calculated at emergency department using routine laboratory test even in remote area may serve as practical and cost-effective marker for guiding the physician in awareness regarding the need for intensive care.

**References**

6. Poudel A, Poudel Y, Adhikari A, Aryal BB, Dangol D,
PMid:33453617

8. Indonesian Society of Respiriology, Indonesian Heart Association, Indonesian Society of Internal Medicine, Indonesian Society of Anesthesiology and Intensive Therapy, Indonesian Pediatric Society, Respirology. 2020;

PMid:32482134

PMid:32334118

PMid:32875598

PMid:32376308

PMid:32302954

PMid:29706967

PMid:33448289

PMid:33244379

PMid:34703296

https://doi.org/10.1177/10760296211010976
PMid:33926262

PMid:32997543

PMid:34437642


PMid:34181675