Correlation between Neutrophile to Lymphocyte Ratio and D-dimer with COVID-19 Severity

Hubungan Rasio Neutrofil Limfosit dan D-Dimer terhadap Derajat Keparahan COVID-19

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ABSTRACT/ABSTRAK

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SARS-CoV-2, a lung-damaging virus, is what causes COVID-19. While a decline in lymphocyte counts suggests immune system harm, a rise in neutrophil counts reflects the severity of the inflammatory response. A high NLR value results from an increase in neutrophils and a decrease in lymphocytes. On the other hand, when plasmin breaks down fibrin to dissolve blood clots, it creates pieces called D-dimers. ARDS is predicted by an elevated D-dimer level. The study's objective was to ascertain how the severity of COVID-19 patients at Dr. H. Abdul Moeloek in 2021 related to NLR and D-dimer levels. Analytical research with a retrospective design is being conducted. The Rank Spearman correlation test and Pearson Chi-Square analyzed medical record data for COVID-19 patients. The study was carried out in Dr. H. Abdul Moeloek in June 2022. 205 COVID-19 patients who underwent NLR and D-dimer testing and were categorized based on the severity of the condition served as the study's subjects. The test findings revealed a substantial connection between NLR and D-dimer (r=0.583), demonstrating a strong and favorable link. NLR and illness severity showed a significant relationship (r=0.49). D-dimer and illness severity also showed a strong connection (r=0.51).

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INTRODUCTION

An international pandemic of the illness known as COVID-19 has been declared. COVID-19 has been a pandemic since March 11, 2020, according to the World Health Organization (WHO) (World Health Organization. 2020). In 2022, there have been 144,121 fatalities from...
COVID-19 in Indonesia alone, while 4,265,187 cases have been verified. (Ministry of Health, 2022). Bandar Lampung City contributed the most, with a total of 11,374 confirmed positive cases of COVID-19 and 798 fatalities in Lampung Province's report of 49,745 persons with COVID-19, which had a death toll of 3,825 people (Dinas Kesehatan Provinsi Lampung, 2022; Dinas Kesehatan Kota Bandar Lampung, 2022).

A virus known as Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) that targets the lungs causes COVID-19 (World Health Organization, 2020). Fever and cough are the significant signs of COVID-19. The majority of COVID-19 patients have little clinical symptoms, while some have a dismal prognosis. Acute respiratory syndrome, severe pneumonia, pulmonary edema, or multiple organ failure symptoms are present in COVID-19 patients who experience deterioration, and these patients finally pass away (Yang et al., 2020).

The NLR is one of the laboratory assessments of the systemic inflammatory response that is typically used to determine the prognosis of COVID-19. The NLR is computed by dividing the number of neutrophil cells per microliter of blood by the number of lymphocyte cells. Compared to patients with a mild prognosis, COVID-19 patients with severe symptoms exhibited higher NLR values (Qin et al., 2020).

The case-control study conducted by Sayed et al. showed that the NLR in the COVID-19 patient group was significantly higher, 2.9 (2.24-3.31) compared to the control group 2.18 (2.3-2.6) with p-value=0.0001 (Sayed et al., 2021). On the other hand, another study conducted by Yang et al. concluded that NLR is a prognostic biomarker that significantly correlates with mortality in COVID-19 patients (Yang et al., 2020). A cohort study conducted at the Zhongnan Hospital of Wuhan University by Yuwei Liu et al. stated an 8% higher risk of death in the hospital for each increase in NLR (r=1.08, p-value=0.0147). In addition, Aijia Ma et al. stated that a high NLR (NLR> 9.8) indicates a higher incidence of ARDS (p-value = 0.005). Therefore, an increase in NLR can be used as a potential risk marker to assess COVID-19 factors (Liu et al., 2020; Zhou et al., 2020; Ma et al., 2020).

On the other hand, D-dimers are pieces created when plasmin breaks down fibrin to dissolve blood clots. A routine D-dimer test confirms a suspected thrombosis diagnosis (Ozen et al., 2021; Yao et al., 2020). Acute Respiratory Distress Syndrome (ARDS), the requirement for care in an intensive care unit, and death are all predictably associated with increased D-dimer levels, frequently detected in severe COVID-19 patients. In COVID-19 patients, a rise in D-dimer >1.0 g/mL predicts mortality (Yao et al., 2020; Zhou et al., 2020). D-dimer levels in the COVID-19 patient group were higher, with a D-dimer level of 966.0 ng/mL, compared to the control group with a D-dimer level of 270.0 ng/mL (p-value 0.001) in Bertolin's case-control study to examine coagulation markers (Bertolin et al., 2021).

According to a study by Wenjing Ye et al., patients who passed away had statistically more significant start and peak levels of D-dimer and NLR than patients who lived (p-value 0.001). The levels of D-dimer and NLR tend to rise noticeably throughout hospitalization in deceased patients, even though they were lower at the initial test than at their peak levels. D-dimer and NLR values were significantly greater in intubated patients than in non-intubated patients at baseline and peak values (p-value 0.001). According to the study's findings, COVID-19 patients' ages and the highest levels of D-dimer are predictive indicators for death. For COVID-19 patients, elevated D-dimer and NLR, as well as test results that are greater than critical values, maybe a warning sign of impending death (Ye et al., 2020).

In light of this context, we intend to research the impact of COVID-19 patients' severity and the Neutrophil Lymphocyte Ratio (NLR) and D-dimer levels.

**METHOD**

This study employs an analytical, retrospective methodology. In order to conduct the study, secondary data on COVID-19 patients who received treatment in Dr. H. Abdul Moeloek's isolation room between July 1 and July 31, 2021, were gathered in June 2022. The research sample consisted of 205 COVID-19 patients who met the age requirement of >18 years old and received treatment in the isolation room at RSUD Dr. H. Abdul Moeloek. These patients completed NLR and D-dimer tests. Exclusion criteria included COVID-19 patients who were postpartum and pregnant, had a history of cardiac disease, were recovering from surgery, had a stroke, Disseminated Intravascular Coagulation (DIC), Deep vein thrombosis (DVT), pulmonary embolism (PE), or had viral infections (HIV, Hepatitis C, Rubella, and others).
Secondary data from the evaluation of the Neutrophil to Lymphocyte Ratio (NLR) and D-dimer levels when measured before patients get any treatments, as well as information on the severity of disease among COVID-19 patients at RSUD Dr. H. Abdul Moeloek Lampung Province, were used in this study. The study was carried out with approval from the ethical committee, and a Certificate of Ethical Eligibility was issued on April 14, 2022, No.046/KEPK-TJK/X/2022.

RESULTS

In July 2021, 205 COVID-19 patients received treatment at Dr. H. Abdul Moeloek's isolation room for this trial. The study participants, 50 female patients (56.2%) and 39 male patients (43.8%) were in the mild severity group. 31 patients (45.6%) were male, and 37 patients (54.4%) were female in the group with moderate severity. In contrast, there were 24 patients (50.0%) who were male and 24 patients (50.0%) who were female in the extreme severity group.

Regarding the age range of the study participants, the age range of 26–45 years had the highest degree of mild severity, represented by a total of 41 patients (46.1%), while the age range >65 years had the lowest, represented by a total of 7 patients (7.9%). The 46-65 year age group had the most patients (61.8%) in the moderate severity category, with a total of 42, and the 18-25 year age group had the fewest (2.9%), with 2 individuals. In contrast, the age group of 18 to 25 years had the fewest patients (2.1%), and the age group of 46 to 65 years had the most patients (50.0%) in the group of severe degrees.

Comparatively, there were 6 patients (6.7%) with concomitant conditions and 83 patients (93.3%) without them in the group of patients with mild severity. 11 patients (16.2%) and 57 patients (83.8%), respectively, in the group with moderate severity did not have any coexisting diseases. In contrast, 12 patients (25.0%) and 36 patients (75.0%), respectively, in the group with severe severity did not have any concomitant diseases. Asthma, dyspepsia, diabetes, and hypertension are comorbid diseases that affect patients.

Table 1. Baseline characteristic of 205 COVID-19 patients

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Degree of COVID-19 Severity</th>
<th>Total (n=205)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mild (n=98)</td>
<td>Moderate (n=68)</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>39</td>
<td>43.8</td>
</tr>
<tr>
<td>Female</td>
<td>50</td>
<td>56.2</td>
</tr>
<tr>
<td>Age Group</td>
<td></td>
<td></td>
</tr>
<tr>
<td>18-25</td>
<td>10</td>
<td>11.2</td>
</tr>
<tr>
<td>26-45</td>
<td>41</td>
<td>46.1</td>
</tr>
<tr>
<td>46-65</td>
<td>31</td>
<td>34.8</td>
</tr>
<tr>
<td>&gt;65</td>
<td>7</td>
<td>7.9</td>
</tr>
<tr>
<td>Comorbid Status</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Without Comorbid</td>
<td>83</td>
<td>93.3</td>
</tr>
<tr>
<td>With Comorbid</td>
<td>6</td>
<td>6.7</td>
</tr>
</tbody>
</table>

The frequency distribution in Table 2's data analysis reveals that the mean and standard deviation (SD) for NLR in the mild severity group are 3.29 and 3.23, respectively. The mean SD for NLR in the group with moderate severity was 5.90±5.15. The mean and standard deviation for NLR in the group with severe severity were 9.84 and 7.78, respectively.

Table 2. NLR and D-dimer Frequency Distribution Depending on Disease Severity in COVID-19 Patients.

<table>
<thead>
<tr>
<th>Degree of COVID-19 Severity</th>
<th>NLR Mean ± SD</th>
<th>D-Dimer Mean ± SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mild</td>
<td>3.29 ± 3.23</td>
<td>841 ± 1083</td>
</tr>
<tr>
<td>Moderate</td>
<td>5.90 ± 5.15</td>
<td>1526 ± 1600</td>
</tr>
<tr>
<td>Severe</td>
<td>9.84 ± 7.78</td>
<td>2541 ± 2192</td>
</tr>
</tbody>
</table>

The mild severity group had a mean and SD of 841 and 1,083 ng/mL in the D-dimer values. D-dimer values in the group with moderate severity were 1,526±1,600 ng/mL on average, standard deviation included. D-dimer levels were 2,541±2,192 ng/mL on average in the group with severe severity, as opposed to that.
Analytical findings demonstrate in Figure 1 a 0.000 (p<0.05) significant association between NLR and D-dimer. The calculated correlation coefficient value is 0.583 using Pearson correlation, which points to a high positive correlation direction. Every increase in the NLR value will be followed by an increase in the D-dimer level, and vice versa, according to the direction of the positive connection.

Figure 1. Scatter Plot Correlation between NLR and D-Dimer in COVID-19 Patients

According to the analysis’s findings using Pearson correlation, depicted in Figure 2, there is a power of r=0.49, which indicates a substantial positive connection between NLR and the degree of disease severity. The same thing can be seen in the findings of the D-Dimer study and disease severity, where there is a substantial positive correlation with a power of r=0.51 and a positive association with illness severity.

Figure 2. Heat Map Correlation between NLR and D-Dimer to The Degree of Disease Severity in COVID-19 Patients

DISCUSSION

The findings of this study suggest that virally induced inflammation may contribute to an increase in NLR. Leukocytes that actively migrate into the immune system or organs primarily comprise neutrophils. Virus-associated inflammatory substances made by lymphocytes and endothelial cells can also stimulate neutrophils. Additionally, systemic inflammation significantly inhibits cellular immunity, which lowers levels of CD4+ T lymphocytes and raises CD8+ suppressor T cells. These changes affect the immunological response to viruses, which mostly rely on lymphocytes. NLR is one sign of a regular inflammatory response frequently used to forecast how patients with viral pneumonia would fare. In COVID-19 patients, elevated NLR can be employed as a separate prognostic biomarker to track the onset of pneumonia (Yang et al., 2020).

Additionally, the findings of this study demonstrate the numerous ways elevated D-dimer levels in COVID-19 patients occur. Primary viral infection results in endothelial damage and a significant increase in the production of cytokines that promote inflammation. The procoagulant state, which can result in pulmonary vascular microthrombosis, respiratory failure, and ARDS, is exacerbated by endothelial injury, hypoxic circumstances, and a persistent inflammatory response (Joly et al., 2020; Connors JM. et al., 2020). In order to prevent a thrombus from forming and clogging blood arteries, plasmin then breaks down the clot made of fibrin and fibrinogen into fibrin(ogen) degradation product (FDP), which is what results in elevated levels of D-dimer in COVID-19 patients (Weitz et al., 2017).

SARS-CoV-2 accumulation in the lungs can activate the immune system by releasing inflammatory cytokines. SARS-CoV-2 causes an immunological reaction when it enters respiratory epithelial cells, causing the release of inflammatory cytokines and a mild interferon (IFN) response. Granulocyte-macrophage colony-stimulating Factor (GM-CSF) and interleukin-6 are examples of pro-inflammatory cytokines that SARS-CoV-2 can immediately release after activating pathogenic Th1 cells (IL-6). Inflammatory monocytes that GM-CSF has activated release many IL-6, tumor necrosis factor-a (TNF-a), and other cytokines. Neutrophils may release extracellular traps known as extracellular NETs, which can cause cytokine release (Hu et al., 2021).
Virus-associated pro-inflammatory cytokines, including those made by lymphocytes and endothelial cells and IL-6, IL-8, tumor necrosis factor, GM-CSF, and interferon-gamma factors, can cause neutrophil discharge. The majority of lymphocytes are responsible for the immune response brought on by viruses, and systemic inflammation dramatically inhibits cellular immunity, causing a considerable decrease in CD4+ T lymphocyte levels and a rise in CD8+ suppressor T cells, which raises NLR values (Yang et al., 2020).

Following this, activated endothelium will be attached to monocytes, neutrophils, platelets, and microparticles in the blood circulation. Tissue Factor (TF) and Neutrophil Extracellular Traps (NETs) will then start coagulation, producing an excessive amount of thrombin and creating a hypercoagulable state (Joly et al., 2020). When plasmin breaks down fibrin to dissolve the blood clot, D-dimer is then formed. Increased blood D-dimer levels can indicate thrombosis suspicion (Yao et al., 2020).

The analysis's findings indicate that the proportion of patients with elevated NLR increases as illness severity increases. According to research by Yang et al. and Fu et al., elevated NLR was a predictive biomarker that independently influences the development of pneumonia in COVID-19 patients. Elevated NLR strongly correlates with illness severity (Yang et al., 2020; Fu et al., 2020). Inflammatory response strength is indicated by an increase in neutrophils, whereas immune system injury is shown by a reduction in lymphocytes. A high NLR value results from a decrease in lymphocytes and an increase in neutrophils. Higher NLR values were seen in COVID-19 individuals with severe and critical symptoms compared to patients with milder symptoms. This suggests a potentially serious condition (Liu et al., 2020; Qin et al., 2020; Ma et al., 2020; Liu et al., 2020). Inflammatory status can thus be predicted using NLR levels. In order to evaluate COVID-19 risk factors, an increase in NLR can, therefore, be employed as a potential marker (Liu et al., 2020).

The other analysis's findings indicate that the proportion of patients with high D-dimer levels increases with the severity of the disease. Similar to the study of Levi et al., which revealed that intensive care unit patients' D-dimer levels were substantially greater (2.4 mg/L) than those of non-ICU patients (0.5 mg/L). D-dimer levels were more significant in COVID-19 patients with severe clinical symptoms than those with mild or moderate symptoms. D-dimer values were higher in patients with severe pneumonia than those with no or mild pneumonia. (Ozen et al., 2021) Levi and colleagues in 2020.

The coagulation process will be triggered by damage to blood vessels caused by various factors, such as inflammation and SARC CoV-2 infection, so the thrombin produced will convert fibrinogen into fibrin monomer. The body uses F XIIIa to cross-link two neighboring "D" domains to create a robust clot to halt bleeding. In order to prevent thrombus, which obstructs blood arteries, the process of fibrinolysis also takes place. Plasmin breaks down the clot made of fibrin and fibrinogen into fibrin(ogen) degradation product (FDP). In order to dissolve blood clots, the hypercoagulable state promotes the production of D-dimers. Because of this, elevated D-dimer correlates with disease severity and serves as a valid predictive indicator for mortality in COVID-19 patients who are hospitalized (Weitz et al., 2017; Yao et al., 2020).

CONCLUSION

According to the findings of this study, NLR and D-Dimer are related to the severity of COVID-19 patients' conditions. In order to develop a therapeutic treatment for COVID-19 infection and lower mortality, additional research is required to examine the components that contribute to D-Dimer and NLR alterations in varying degrees of severity.

REFERENCES


