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Major Inflammatory Markers and Their Significance in Predicting Severity of COVID-19 Disease Pattern

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Abstract

The unanticipated outbreak of the COVID-19 pandemic has shocked the world in terms of both lives and livelihood. SARS-CoV-2 virus primarily affects the respiratory system, although other organ systems are also involved. Early diagnosis followed up by a retrospective analysis and tracking of a few markers relevant to the immunological status of the individual may aid in determining the state of the patient's disease prognosis. The aim of the present study was to evaluate immunological parameters such as neutrophil to lymphocyte ratio (NLR), C-reactive protein (CRP), and D-dimer, taking into account the patient's age and oxygen saturation level.

Our retrospective analysis of clinical data revealed that such parameters as CRP, D-dimer, and NLR should be taken into consideration to predict severe COVID-19-related complications. The data obtained indicate that patients over age 60 are especially vulnerable to severe COVID-19.(**International Journal of Biomedicine. 2021;11(4):488-492.**)

Key Words: COVID-19 • neutrophilia • D-dimer • C-reactive protein

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Introduction

The unanticipated outbreak of the COVID-19 pandemic has shocked the world in terms of both lives and livelihood. The origin of the SARS-CoV-2 virus is itself a debatable mystery; however, its severity is highly unpredictable, as several countries in the world have already experienced serious consecutive waves of infection. As the virus mutates, it becomes more infectious and aggressively hits the host, leading to a higher rate of mortality.[1] Globally, this virus has been considered a major threat in both developed and under-developed countries as it mainly halts advancement of the socio-economic status of the country directly.

SARS-CoV-2 establishes residence in the host and multiplies, primarily focusing on the respiratory system and causing a respiratory disease. It enters the epithelial cells of the nasal cavity by engaging the ACE2 receptor with the viral receptor-binding domain (RBD) and begins replicating.[2-4] In the stage of established pulmonary disease, there are viral multiplication and localized inflammation in the lungs that may lead to devastating damage. The most severe stage of the illness manifests as an extrapulmonary, systemic hyperinflammation syndrome.[5]

It is highly important to block the viral entry at its initial phase. Therefore, early diagnosis and monitoring are necessary in order to control the disease progression and its clinical complications.[6] Early diagnosis followed by the retrospective analysis and follow-up of few markers could even predict

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the immunological status of the individual. The heightened immunological response and the consecutive transmission of this virus colonizing in the lungs are the two critical factors that determine the severity of the disease.[7]

Miriam Merad and Jerome C. Martin highlight that current models of COVID-19 propose three distinct immune stages that are crucial for the ultimate disease course:[8] "In the first stage, early activation of the immune system through the induction of a potent interferon response is important to control the virus. In the second stage, a delayed interferon response may lead to progressive tissue damage. This may ultimately lead to the third stage, a deleterious hyperinflammation characterized by the excessive macrophage activation and coagulation that is seen in patients with severe disease, possibly followed by dysregulation of tissue repair mechanisms and fibrosis."

According to Huertas et al., endothelial cell dysfunction and impaired microcirculatory function contribute markedly to life-threatening complications of COVID-19, such as venous thromboembolic disease and multiple organ involvement.[9] Endothelial dysfunction, complement activation, thrombin generation, platelet and leukocyte recruitment, and the initiation of innate and adaptive immune responses culminate in immunothrombosis, ultimately causing (micro)thrombotic complications, such as deep vein thrombosis, pulmonary embolism, and stroke. In this regard, the activation of coagulation and thrombocytopenia has emerged as a prognostic marker in COVID-19.[10,11]

Coagulopathy and D-dimer elevations were seen in 3.75%–68.0% of the COVID-19 patients.[12,13] Yao et al.[14] showed that a D-dimer level of >2.14 mg/L predicted in-hospital mortality in COVID-19 patients with a sensitivity of 88.2% and specificity of 71.3% (AUC=0.85; 95%CI=0.77-0.92).[14] High levels of CRP have also been used as an indicator of COVID-19 disease severity. Stringer et al. showed that a threshold cut-off of CRP ≥ 40 mg/L was associated with mortality in COVID -19 patients.[15]

As COVID-19 progresses to the lethal final phase, it is necessary to monitor the surge levels of inflammatory markers in order to prevent the development of cytokine storm and further complications. The aim of the present study was to evaluate immunological parameters such as neutrophil to lymphocyte ratio (NLR), C-reactive protein (CRP), and D-dimer, taking into account the patient's age and oxygen saturation level.

Materials and Methods

The study was investigated at Najran Armed Forces Hospital. Patients suspected of having COVID-19 were confirmed by qRT-PCR analyzing the fold change expression of SARS-CoV-2 viral-specific genes, such as the *N* gene and *E* gene in an oro-nasal throat swab. Strong COVID-19 positive patients with qRT-PCR cycle threshold values of 20-25 consented to enroll in this study. Nearly 121 strong positives volunteered for this clinical investigation. This study included the patients' complete data from the date of admission until the date of discharge or death, information that was available from the hospital administrative records.

We analyzed plasma NLR, CRP, and D-dimer values. All these parameters are closely correlated with immune status and internal microvasculature clotting, which is associated with the lethal phase of the disease. This basic diagnostic profile is usual for the typical multi-specialty hospital.

The case files of the patients were retrospectively reviewed, the required information was extracted, and patients with a co-morbidity were excluded. Treatment with azithromycin, hydroxychloroquine, lopinavir-ritonavir, steroids, or oxygen support was considered as dependent upon the patients' requirements. Asymptomatic patients, were given multivitamins and zinc tablets. The sample size was determined by the time window of the study.

Statistical analysis was performed using the IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.).

Results

A scatter plot (Fig.1) shows a moderate positive linear relationship between NLR and patient's age. The COVID-19 patients above 60 were more sensitive to increased NLR levels. We found a moderate positive linear relationship between CRP and patient's age (Fig.2).

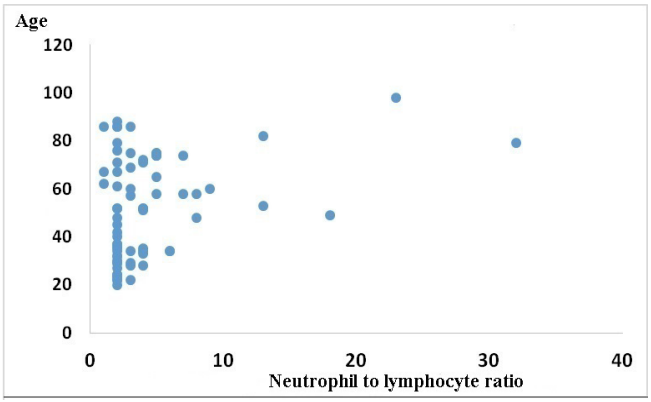

Fig. 1. *The relationship between NLR and patient's age (n=121).*

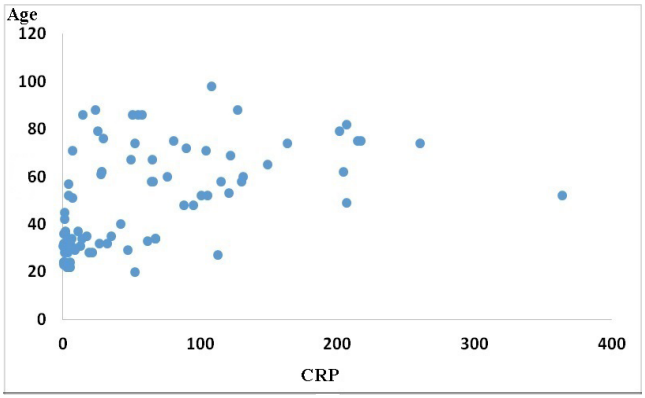

Fig. 2. *The relationship between CRP and patient's age (n=121).*

The COVID-19 patients above 60 developed higher CRP values. Figure 3 shows a moderate positive linear relationship between D-dimer and patient's age. The COVID-19 patients above 60 were more sensitive to increased D-dimer values.

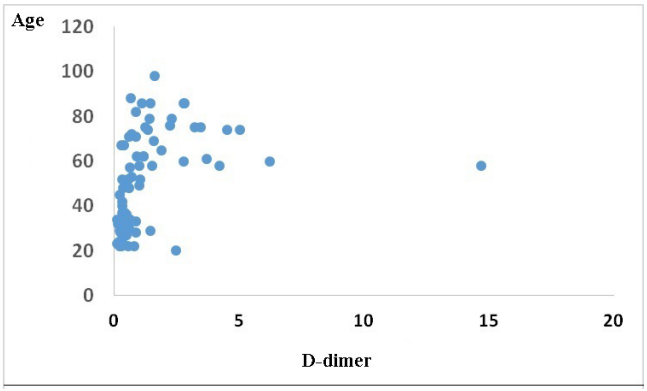

Fig. 3. *The relationship between D-dimer and patient's age (n=121).*

Figure 4 shows a strong negative linear relationship between patient's age and SaO_2: SaO_2 levels decrease in patients above 60 years. In contrast, the age groups ranging between 20-40 and 40-60 maintain consistent SaO_2 levels.

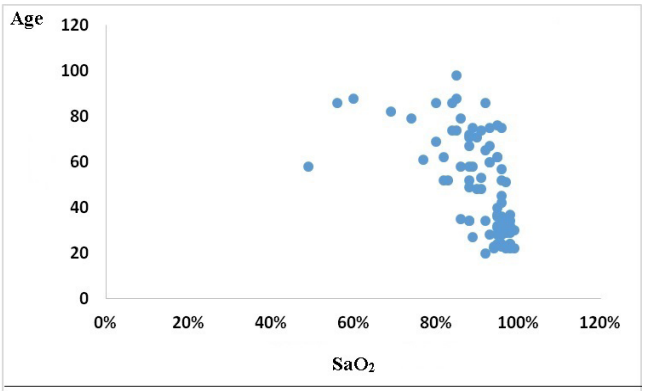

Fig. 4. *The relationship between patient's age and SaO_2 (n=121).*

Discussion

COVID-19 has quickly spread around the world with high mortality worldwide. The pathological pulmonary damage it has caused may be directly related to the viral destruction of alveolar and bronchial epithelial cells or mass production of proinflammatory cytokines (cytokine storm).[16] In COVID-19, the cytokine storm may result in an uncontrolled systemic inflammatory response, ARDS, multiple organ failure, and death in severe cases.[17,18]

Currently, it is clear that hyperinflammation and coagulopathy contribute to disease severity and death in patients infected with SARS-CoV-2.[8] It is believed that higher values of proinflammatory markers are related to extensive lung injury.[19] The neutrophils are known to develop a sophisticated network of extracellular fibers composed of DNA containing histones, called neutrophil extracellular traps (NETs). There is some evidence to suggest that NETosis is conditional on the production of reactive oxygen species.[20]

It has been shown that neutrophilia predicts a poor outcome in patients with severe COVID-19 cases, and NLR may be an independent risk factor for the severity of this disease.[21] In a recent article, Shivakumar, with associates from India, found that the neutrophil-to-lymphocyte-to-monocyte ratio and the platelet-to-lymphocyte ratio were significantly prognostic in COVID-19.[22]

The high mortality associated with thromboembolic disorders in COVID-19 has prompted clinicians to use D-dimer as a useful marker for assessing the severity of the disease.[23] According to a review by Harvard Medical School researchers, in critically ill patients with COVID-19, elevated levels of D-dimer were found in 100% of participants, elevated fibrinogen in 74%, and factor V11 in 100%.[24]

Nalbant et al.[25] found that the risk of COVID-19 was 20.3-fold greater when NLR was ≥2.4 in the logistic regression (P=0.007). The authors concluded that NLR is an independent predictor for the diagnosis of COVID-19. In a study by Seyit et al.,[26] the CRP (P=0.0001) and NLR (P=0.001) remained significantly higher in the patients with positive SARS-CoV-2 PCR test results.

The biological changes linked to aging and morbidity are one of the reasons deaths have been concentrated among older persons around the world. In addition to having less ability to fight off a novel virus, several aspects of immune functioning also may be worse for older people. Hyperinflammation has been linked to poor outcomes with COVID-19 due to "cytokine storms," or an out-of-control immune reaction. The average number of dysregulated cytokines doubles in the age group from the 50s to the 80s.[27]

In our analysis, this NLR ratio was observed to be more in cases of patients belonging to the age group of 60 and above. Our analysis was in agreement with the previously reported data; therefore, it is well understood that not only monitoring the immunological profiles but also the patient's age should be taken into consideration in the case of COVID-19.

Conclusion

Our retrospective analysis of clinical data revealed that such parameters as CRP, D-dimer, and NLR should be taken into consideration to predict severe COVID-19-related complications. The data obtained indicate that patients over age 60 are especially vulnerable to severe COVID-19.

Competing Interests

The authors declare that they have no competing interests.

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