Original Research Article

D-dimer levels and lymphocyte counts as prognostic and predictive factors in children with COVID 19

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Abstract: Background: Coronavirus Disease 2019 (COVID-19) is characterized by high fever, sudden developing respiratory distress. The Purpose of this study is to identify the association of D-dimer levels and lymphocyte counts with poor prognosis and to predict the clinical course in patients with COVID-19.
Methods: A Total of 85 hospitalized children diagnosed with COVID 19 were included in the study. According to AIIMS guidelines, they were divided into two groups, one with severe disease (N=23) and other with non-severe disease which included mild to moderate cases (n=62), distinctive performance analysis of these values were performed and the cut off values were determined.
Results: A total of 85 hospitalized patients with COVID-19 were included in the final analysis. The mean age was 10 ± 5 years and 45 (52.9%) were males. Lymphocyte count was found as statistically significantly low (p <0.001) while D-dimer level was statistically significantly higher in the group with severe disease (p <0.001). As for the effectiveness of lymphocyte count in distinguishing severe and non-severe patients with COVID-19 when cut-off score 1500/mm3 was taken, sensitivity was 30% and specificity was 77% and that of D-dimer when cut-off score 2 mg/L was taken, sensitivity was 22% and specificity was 50%. D-dimer level was found to have a significant discrimination power (AUC = 0.879, p < 0.0001,95% CI).
Conclusions: The lymphocyte value of $\leq 1500/mm3$ and D-dimer value of $\geq 2mg/L$ can be used in the early determination of patients with poor prognosis in COVID-19. Using these cut-off values for D-dimer and lymphocyte count will help predict prognosis and make rapid treatment decisions in patients with COVID-19.

1. Introduction

A novel coronavirus-related pneumonia case was discovered in Wuhan, China, in December 2019. As a result, an epidemic spread throughout China, followed by an increase in cases in other countries. The Coronavirus Working Group of the International Virus Taxonomy Committee proposed naming this virus severe acute respiratory syndrome coronavirus 2. (SARS-CoV-2)[1].

High fever, normal or decreased white blood cell count, lymphopenia, sudden onset respiratory distress, and radiological findings are all symptoms of the disease [2]. The World Health Organization (WHO) declared the epidemic a global emergency on January 30, 2020 [3].

Pediatric age groups account for 1%-5% of COVID-19 diagnoses, with less than 1% under the age of 10. COVID-19 appears to affect children less severely than adults. According to the diagnosis, 90% of paediatric patients were asymptomatic, mild, or moderately ill. However, 6.7% of cases may be severe [4].

Children may be a critical target population for effective epidemic-reduction methods because they may play a significant role in the dynamics of pathogen transmission and outbreaks [5].

Children may have been underrepresented because they are more likely than adults to have asymptomatic or mild infections and have better overall outcomes. Cough, fever, and exhaustion are the most common symptoms in children, but atypical symptoms such as vomiting and diarrhoea are also common; anosmia

and ageusia are usually mentioned in pre-teenagers and adolescents because younger children are sometimes unable to explain these types of symptoms [6].

While children's symptoms are milder than those of adults, severe disease can still occur, particularly in children with comorbidities. Paediatric data on the inflammatory markers that influence disease prognosis are limited [7].

To determine severity, laboratory indicators, as well as clinical manifestations, must be evaluated. D-dimers and C-reactive protein levels rise in patients infected with SARS-CoV-2 [8].

Lymphocytopenia was linked to a poorer outcome in COVID-19 patients, and increased D-dimers were also associated with a poor prognosis in critically ill COVID-19 patients in an adult study [9].

COVID-19 is frequently complicated by coagulopathy, and disseminated intravascular coagulation (DIC) is found in the majority of deaths. Coagulation test abnormalities in SARS-CoV-2 infected patients are expected to develop as a result of the severe inflammatory response. In particular, an unusual fever and hyperinflammatory process has emerged in COVID19 paediatric populations [10].

Severe COVID-19 is often complicated by coagulopathy, and disseminated intravascular coagulation (DIC) is detected in most deaths [11]. Critically ill COVID-19 patients frequently develop coagulopathy. A recent autopsy confirmed that the majority of fatalities were caused by systemic microvascular thrombosis. However, the role of the coagulation parameter D-dimer in the development of COVID-19 is less clear. The most common cause of an increase in D-dimer levels is pulmonary thrombosis [12].

2. Methods

A Total of 85 hospitalised children diagnosed with COVID-19 were included in the study. Out of which, 40 were female, and 45 were male. The study was undertaken at KIMS Hospital, Bangalore. The duration of the study was from August 2020 to May 2021.

Patients from 6 months to 17 years were considered. Those diagnosed with COVID-19 were included in the study (laboratory-confirmed cases, SARS-CoV-2 RNA detected by molecular method). Patients with comorbid diseases like nephrotic syndrome, type-1 DM, or those receiving corticosteroid treatment were excluded from the study.

According to the AIIMS PAEDIATRIC COVID-19 Treatment Guidelines, patients with COVID-19 were divided into two groups as patients with severe disease (1- Hypoxia presence: Oxygen saturation $\leq$ 93% on room air, respiratory distress. 3- lung infiltrates > 50% on chest imaging) and with non-severe disease (mild or moderate).

Considering this classification, 23 patients were included in the severe COVID-19 group, while 62 were included in the non-severe group. D-dimer level and lymphocyte count values were compared between severe and non-severe COVID-19 patient groups.

Distinctive performance analysis of the values significantly different between the groups was performed, and cut-off values were determined.

3. Results

A total of 85 hospitalized patients with COVID-19 were included in the final analysis. The mean age was 10 $\pm$ 5 years and 45 (52.9 %) were males and 47.1% were females. There was no statistically significant difference between severe COVID-19 patient group (n= 23, 27.1 %) and non-severe COVID-19 patient group (n=62, 72.9 %) in terms of age (p= 0.842). (Figure 1).
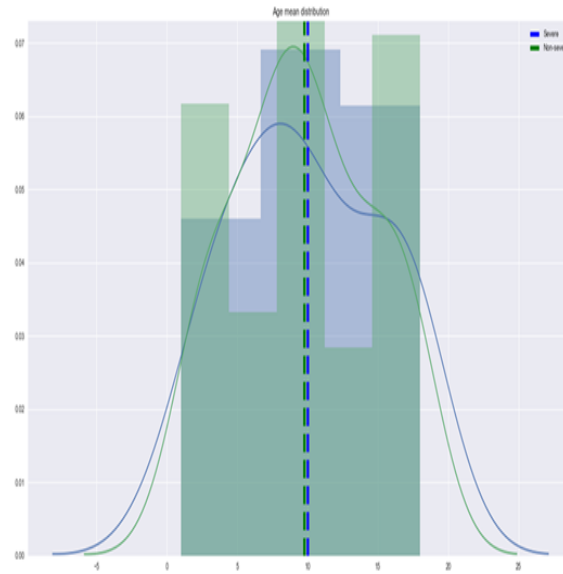
Figure 1. PRISMA flow diagram

Independent sample t-test was performed. ROC analysis of lymphocyte and D-dimer baseline values; AUC (Area under the curve) analysis performed.

Lymphocyte count was found as statistically significantly low (p <0.001) in the severe COVID-19 group while D-dimer level was statistically significantly higher in the group with severe disease (p <0.001).As for the effectiveness of lymphocyte count in distinguishing severe and non-severe patients with COVID-19 when cut-off score 1500/mm3 was taken, sensitivity was 30% and specificity was 77%. (Figure 2).
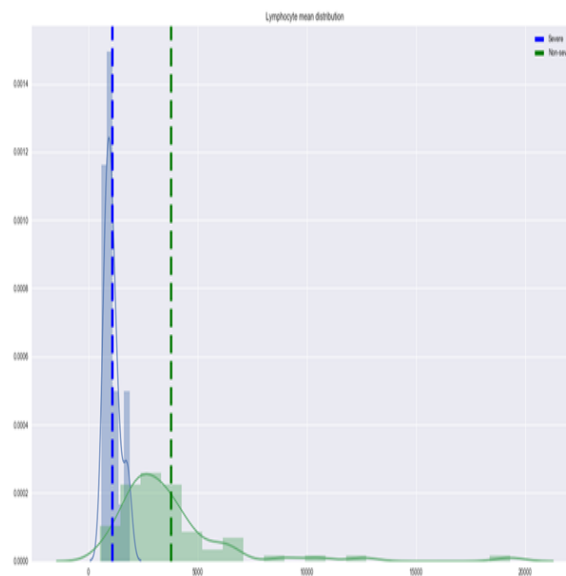

Figure 2. PRISMA flow diagram

As for the effectiveness of D-dimer level in distinguishing severe and non-severe patients with COVID-19 when cut-off score 2 mg/L was taken, sensitivity was 22% and specificity was 50%. (Figure 3).
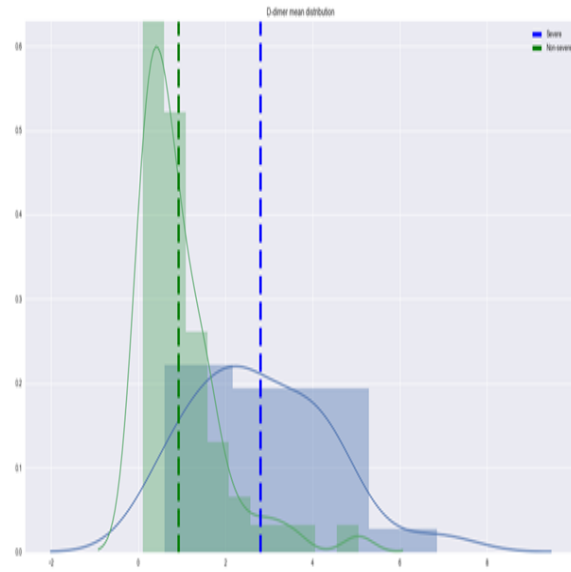
Figure 3. PRISMA flow diagram

Lymphocyte count was found to have a significant discrimination power (AUC = 0.052, p>0.05, 0.95% Confidence interval) . Sensitivity was 30% and Specificity was 77%. (Figure 4)
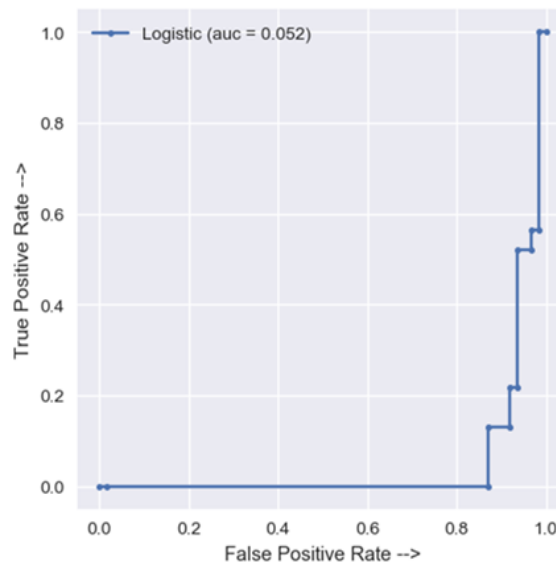

Figure 4. PRISMA flow diagram

D-dimer level was found to have a significant discrimination power (AUC = 0.879, p < 0.0001,95% CI) Sensitivity was 22% and Specificity was 50%. (Figure 5)
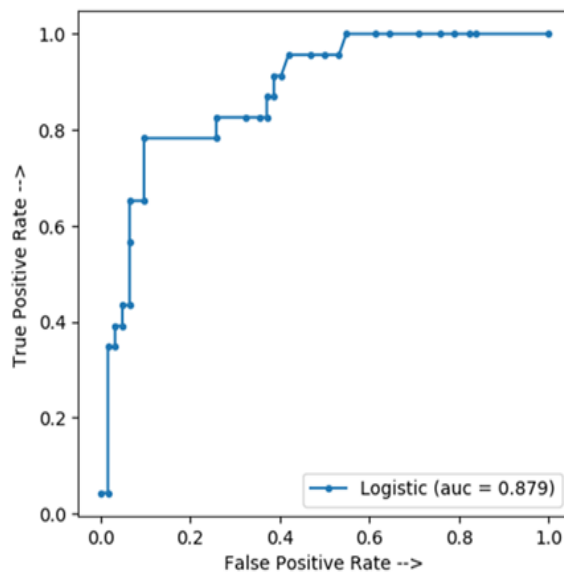
Figure 5. PRISMA flow diagram

4. Discussion

The current study found that patients with severe COVID-19 had a lower lymphocyte count and a higher D-dimer level than patients with non-severe COVID-19. D-dimer is a byproduct of plasmin's enzymatic breakdown of cross-linked fibrin. On the other hand, high D-dimer levels are common in patients suffering from various acute infectious and inflammatory diseases [13]. Endothelial dysfunction causes high levels of D-dimer, thrombin, and fibrin degradation products, thrombocytopenia, and prolonged clotting times, contributing to hypoxia and pulmonary congestion caused by thrombosis and microvascular occlusions [14].

According to recent research, high D-dimer levels correlate with the severity of COVID-19 [15].

D-dimer levels detect thrombosis in patients. In the early stages of COVID-19 disease, studies have shown an increase in D-dimer and fibrinogen concentrations. A 3 to 4-fold rise in D-dimer levels is associated with a bad prognosis [16].

The predominant coagulation abnormalities in COVID-19 patients progress with hypercoagulation, and the risk of venous thromboembolism increases uncontrollably. Some experts call this condition thrombo-inflammation or COVID-19-associated coagulopathy (CAC). In severely affected patients, disseminated intravascular coagulation (DIC) has been reported [17]. Lymphopenia, also known as lymphocytopenia, is a condition characterized by low lymphocyte counts in the blood. Although T cells were initially elevated at the onset of COVID-19, these patients tended to have low lymphocyte counts, associated with increased COVID-19 severity [18].

Multiple mechanisms are involved in causing Lymphopenia. SARSCoV2 may attack lymphocytes directly or destroy lymphoid organs. Indeed, because patients with the severe COVID-19 phenotype have elevated blood lactic acid levels, Lymphopenia could be caused by such metabolic molecules [19]. The lymphocyte count was found to be significantly lower in COVID-19 patients who were followed in the intensive care unit compared to those who were followed in other clinical services [20].

Wang et al. conducted a study on Lymphopenia found in approximately 60% of all patients and 81.5% of those who died. It was concluded that the degree of Lymphopenia could predict the prognosis by indicating the severity of the SARS-CoV-2 invasion or the state of antiviral immunity [21]. Lymphocyte apoptosis is regulated by pro and anti-apoptotic mechanisms mediated by endogenous and exogenous factors [22]. T lymphocyte apoptosis has been linked to Lymphopenia in SARS-CoV and MERS-CoV infections [23].

The cytopathic effect caused by direct infection of T cells is highlighted in SARS-CoV-2 infection. Other predicted lymphopenia mechanisms include pulmonary sequestration during extensive bilateral pneumonia and bone marrow suppression during cytokine storm [24]. A study examining the relationship between D-dimer levels and mortality in COVID-19 patients found that D-dimer levels higher than 2.0 g/mL were independent predictors of mortality at the time of admission [25].

This increase in D-dimer in COVID-19 patients is thought to be due to increased systemic pro-inflammatory activation, which triggers the prothrombotic process. Endothelial injury, complement activation, or other procedures are thought to be directly related to hypercoagulability [26]. There are some limitations to this study. The first is that only the values of the laboratory parameters examined during the application were considered, and changes in the following days were not taken into account.

The values of $\leq 1500/mm3$ lymphocyte and $\geq 2mg/L$ D-dimer can be used in the early determination of patients with good and poor prognosis in COVID-19. As a result, it helps in determining patients with poor prognoses in the early stages and rapid treatment decisions; the morbidity and mortality of these patients will be reduced.

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Conflicts of Interest: "The authors declare that they do not have any competing interests."

References

[1] Gorbalenya AE, Baker SC, Baric RS, Groot RJ, Drosten C, Gulyaeva AA et al. The species Severe acute respiratory syndrome-related coronavirus: classifying 2019-nCoV and naming it SARS-CoV-2. Nat Microbiol. 2020;(5), 536-44.

[2] Recent studies have shown that high D-dimer levels correlate with the severity of COVID-19.

[3] Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M et al.The third international consensus definitions for sepsis and septic shock (Sepsis-3). JAMA 2016;315(8):801-10.

[4] Tezer H, Demirda? TB. Novel coronavirus disease (COVID-19) in children. Turkish journal of medical sciences. 2020;50(9):592-603.

[5] Rajapakse N, Dixit D. Human and novel coronavirus infections in children: a review. Paediatrics and international child health. 2021 Jan 2;41(1):36-55.

[6] Luxi N, Giovanazzi A, Capuano A, Crisafulli S, Cutroneo PM, Fantini MP, Ferrajolo C, Moretti U, Poluzzi E, Raschi E, Ravaldi C. COVID-19 vaccination in pregnancy, paediatrics, immunocompromised patients, and persons with history of allergy or prior SARS-CoV-2 infection: overview of current recommendations and pre-and post-marketing evidence for vaccine efficacy and safety. Drug safety. 2021 Dec;44(12):1247-69.

[7] Zhu F, Ang JY. COVID-19 infection in children: diagnosis and management. Current infectious disease reports. 2022 Apr;24(4):51-62.

[8] Alkan G, Sert A, Emiroglu M, Tuter Oz SK, Vatansev H. Evaluation of hematological parameters and inflammatory markers in children with COVID-19. Irish Journal of Medical Science (1971-). 2021 Sep.

[9] Alharthy A, Aletreby W, Faqihi F, Balhamar A, Alaklobi F, Alanezi K, Jaganathan P, Tamim H, Alqahtani SA, Karakitsos D, Memish ZA. Clinical characteristics and predictors of 28-day mortality in 352 critically ill patients with COVID-19: a retrospective study. Journal of epidemiology and global health. 2021 Mar;11(1):98.

[10] Chiotos K, Bassiri H, Behrens EM, et al. Multisystem inflammatory syndrome in children during the coronavirus 2019 pandemic: a case series. J Pediatr Infect Dis Soc. 2020;9(3):393?398

[11] Connors JM, Levy JH. COVID-19 and its implications for thrombosis and anticoagulation. Blood 2020;135(23):2033-40.

[12] Li Y, Zhao K, Wei H, Chen W, Wang W, Jia L, Liu Q, Zhang J, Shan T, Peng Z, Liu Y. Dynamic relationship between D?dimer and COVID?19 severity. British journal of haematology. 2020 Jul;190(1):e24.

[13] Panigada M, Bottino N, Tagliabue P, Grasselli G, Novembrino C, Chantarangkul V et al. Hypercoagulability of COVID?19 patients in ICU. A report of thromboelastography findings and other parameters of hemostasis. J ThrombHaemos. 2020;18:1738-42.

[14] Connors JM, Levy JH. COVID-19 and its implications for thrombosis and anticoagulation. Blood. 2020 Jun 4;135(23):2033-40.

[15] Li Y, Zhao K, Wei H, Chen W, Wang W, Jia L et al. Dynamic relationship between D?dimer and COVID?19 severity. Br J Haematol. 2020;190:24-7.

[16] Rostami M. Mansouritorghabeh H. d-Dimer level in COVID-19 infection: a systematic review. Expert Rev Hematol. 2020;13(11):1265-75.

[17] Connors JM, Levy JH. Thromboinflammation and the hypercoagulability of COVID?19. J ThrombHaemost. 2020;18:1559-61.

[18] Yang X, Yu Y, Xu J, Shu H, Liu H, Wu Y, Zhang L, Yu Z, Fang M, Yu T, Wang Y. Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. The lancet respiratory medicine. 2020 May 1;8(5):475-81.

[19] Tan L, Wang Q, Zhang D, Ding J, Huang Q, Tang YQ, Wang Q, Miao H. Lymphopenia predicts disease severity of COVID-19: a descriptive and predictive study. Signal transduction and targeted therapy. 2020 Mar 27;5(1):33.

[20] Wang L, He W, Yu X, Hu D, Bao M, Liu H et al. Coronavirus disease 2019 in elderly patients: Characteristics and prognostic factors based on 4-week follow-up. J Infect. 2020;80:639-45.

[21] Huang C, Wang Y, Li X,Ren L, Zhao J, Hu Y et al. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020;395(10223):497-506.

[22] Aksoy E, Azkur AK. Schmallenberg virus induces apoptosis in Vero cell line via extrinsic and intrinsic pathways in a time and dose dependent manner. J Vet Med Sci. 2019 Feb 9;81(2):204-12.

[23] Tan L, Wang Q, Zhang D, Ding J, Huang Q, Tang YQ et al. Lymphopenia predicts disease severity of COVID-19: a descriptive and predictive study. Signal Transduct Target Ther. 2020;5(1):1-3.

[24] Azkur AK, Akdis M, Azkur D, Sokolowska M, Veen W, Bruggen M-C et al. Immune response to SARS?CoV?2 and mechanisms of immunopathological changes in COVID?19. Allergy 2020;75:1564-81.

[25] Zhang L, Yan X, Fan Q, Liu H, Liu X, Liu Z et al. D?dimer levels on admission to predict in?hospital mortality in patients with Covid?19. J Thromb Haemost. 2020;18(6):1324-9.

[26] Magro C, Mulvey JJ, Berlin D, Nuovo G, Salvatore S, Harp J et al. Complement associated microvascular injury and thrombosis in the pathogenesis of severe COVID-19 infection: a report of five cases. Transl Res. 2020;220:1-13.