Cross-sectional Study

Role of hematological parameters in the stratification of COVID-19 disease severity

Sadia Taj, Ambreen Kashif, Syeda Arzinda Fatima, Sheharbano Imran, Ayaz Lone, Qasim Ahmed

FMH College of Medicine and Dentistry Shadman, Lahore, Pakistan

ARTICLE INFO

Keywords:
- Covid-19 disease
- Neutrophil to lymphocyte ratio
- Hematological manifestations

ABSTRACT

Objective: COVID-19 virus involves respiratory as well as other body systems including cardiovascular, gastrointestinal, neurological, immunological and hematopoietic system. Patient of covid-19 pneumonia presents with wide range of hemostatic abnormalities. These hemostatic abnormalities in COVID-19 are related with disease progression, severity and mortality. The Objective of our study is to evaluate the role of hematological parameters in determination of COVID-19 disease severity.

Material and method: This was a retrospective study, conducted in Department of Pathology and Department of medicine, FMH college of Medicine and Dentistry from May 2020 to July 2020. Total of 101, confirmed cases of covid-19 disease, both genders between 17 and 75-year age were included. Hematological parameters were compared in mild, moderate, severe and critical disease group. Continuous variables were analyzed by using non parametric, Kruskal Wallis test while categorical variables were analyzed by chi-square test.

Results: Out of 101 patients, 20.8%, 51.8%, 19.8% and 7.9% were in mild, moderate, severe and critical group respectively. Median (IQR) values of WBCs (p-value 0.004), ANC (p-value 0.002), NLR (p-value 0.001), D-dimer level (p-value 0.001), ferritin (0.0001), LDH (0.0001) were significantly increased in patients with critical disease. Median (IQR) values of APTT (p-value 0.003) and CRP (p-value 0.001) were suggestively higher in patients with severe disease. Other parameters like Hemoglobin, MCV, HCT, ALC, Platelet count, prothrombin time did not show statistically significant association with severity of disease.

Conclusion: The study concluded that Leukocytosis, neutrophilia, elevated Neutrophil to lymphocyte ratio, APTT, D-dimer, LDH and serum ferritin and CRP are associated with severity of covid-19 disease.

1. Introduction

Coronavirus disease causing severe acute respiratory syndrome has rapidly evolved into a global pandemic effecting more than 1 million individuals worldwide [1]. Although, primarily it was documented as a respiratory tract infection, emerging researches indicate that covid-19 causes an illness which has a wide variety of clinical features, ranging from mild to moderate upper respiratory tract infection to severe systemic disease which involves respiratory as well as other body systems including cardiovascular, gastrointestinal, neurological, immunological and hematopoietic system [2,3]

Patients with clinical symptoms, progress to pneumonia frequently with radiological evidence of parenchymal disease. Most of the patients 80.9% present with mild disease, 13.8% with severe and 4.7% with critical disease [4]. Patients admitted to intensive care units manifest high plasma levels of proinflammatory cytokines including interleukins and tumor necrosis factor-α, which suggests that individuals with severe disease may be develop cytokine storm effect [5]. Patients may develop acute respiratory distress syndrome immediately after onset of disease, therefore, there is a great need to diagnose COVID-19 and determine disease severity as early as possible.

The association of hematological abnormalities in severe COVID-19 pneumonia is multifactorial. Hematological abnormalities in COVID-19 are related with disease progression, severity and mortality. Lymphopenia, thrombocytopenia, abnormal coagulation profile and sepsis leading to disseminated intravascular coagulation (DIC) is very well documented in patients of COVID-19 [6]. Platelet count is a simple and effortlessly available hematological parameter, which is independently

* Corresponding author.

E-mail addresses: sadia.taj@fmhcmd.edu.pk (S. Taj), ambreen.kashif@fmhcmd.edu.pk (A. Kashif), arzinda.fatima@fmhcmd.edu.pk (S. Arzinda Fatima), shehar.imran@fmhcmd.edu.pk (S. Imran), ayaz.lone@fmhcmd.edu.pk (A. Lone), qasim.ahmed@fmhcmd.edu.pk (Q. Ahmed).

https://doi.org/10.1016/j.amsu.2020.12.035

Received 24 November 2020; Received in revised form 16 December 2020; Accepted 20 December 2020

Available online 8 January 2021

2049-0801/© 2021 The Authors. Published by Elsevier Ltd on behalf of IJS Publishing Group Ltd. This is an open access article under the CC BY-NC-ND license
associated with disease severity and risk of mortality in the intensive care unit (ICU) [7].

Coagulopathies like disseminated intravascular coagulation, sepsis-induced coagulopathy (SIC), local microthrombi, venous thromboembolism (VTE), arterial thrombotic complications, and thrombo-inflammation have been associated with COVID-19 [8]. Hematological manifestations of this particular virus should be tracked closely as this epidemic evolves.

These hematological and inflammatory biomarkers like Complete Blood Count, d-dimer, C-Reactive Protein, Ferritin and coagulation profile can play a vital role in early prediction of disease severity and can provide a better guide for prompt management of patients, thus, can help in deceasing the disease morbidity and mortality.

In this study, we aim to investigate the association of hematological parameters with COVID-19 disease, and to evaluate the role of hematological parameters in stratification of COVID-19 disease severity.

2. Materials and methods

This was a retrospective cross-sectional study, conducted in Department of Pathology and Department of Medicine, FMH college of Medicine and Dentistry. Data was recruited after taking approval form Institutional Review Board (IRB# FMH-08-2020-IRB-776-M). Data of three months May 2020 to July 2020 was included. Total of 101 patients, both genders between 17 and 75-year age were studied. Patients with known case of chronic liver disease and known hematological diseases were excluded. Patients were categorized into mild, moderate, severe and critical disease according to their clinical features.

Complete Blood Count was performed on Sysmex XN-10, coagulation profile was performed on CA-5500 while lactate dehydrogenase (LDH) and C-reactive protein (CRP) were performed on Roche Cobas c311 and serum Ferritin was performed on Abbott Architect I 1000 SR.

Mild disease was defined as symptoms of fever, sore throat, cough and no sign of pneumonia on X-rays. Moderate disease was defined as fever and respiratory symptoms with radiological imaging of less than 50% lung involvement and oxygen saturation <93%. Patients with respiratory distress (respiratory rate >30 breath per min, O2 saturation less than 93% and more than 50% lung infiltrate were classified as severe disease while patients with respiratory failure requiring mechanical ventilation, shock or other organ failure were classified as patients having critical disease.

2.1. Statistics

Descriptive analysis was performed on all the variables. Data was stratified according to age, gender, and severity of disease. Hematological and coagulation parameters Laboratory parameters were compared between mild, moderate, severe and critical disease. Quantitative variables white blood count (WBC), absolute lymphocyte count (ALC), absolute neutrophil count (ANC), neutrophil to lymphocyte ratio (NLR), hemoglobin (Hb), mean corpuscular volume (MCV), Platelet count, mean platelet volume (MPV), Prothrombin time PT, activated partial thromboplastin time aPTT, Ferritin, D-dimer, lactate dehydrogenase (LDH), C-reactive protein (CRP) were analyzed by Median (IQR) whereas Categorical variables like gender, temperature, Oxygen saturation, Oxygen requirement and lung infiltrates were presented in form of frequency and percentage. Continuous variables were analyzed by using non parametric, Kruskal Wallis test while categorical variables were analyzed by chi-square test. Data was analyzed in SPSS version 25. P-value equal or less than 0.05 was considered significant.

3. Result

Out of 101 patients, 66(65.3%) were males and 35(34.7%) were females. Patients with mild and moderate disease were younger as compared to patients present with severe and critical disease. The median (IQR) age of the patients in mild, moderate, severe and critical patients were 45(32), 58(28), 63(16) and 59(22) years respectively. The covid-19 disease was more prevalent in males 66(65.3%) as compared to females 35(34.7%) (p value 0.495) and male patients had more severe disease as compared to females. Demographic data and clinical findings of patients are shown in Table 1.

Median (IQR) respiratory rates were highest in patients with critical disease 35(4) per minute (p-value 0.0001) compared to other groups. Among 101 patients, 97(96%) patients have raised body temperature. Oxygen saturation was less than 93% in 58 (57.4%) while it was more than 93% in 43(42.6%) patients. (p-value 0.0001). Median (IQR) of O2 saturation was 97(2)%, 91(9) %, 86(10) %, 82(25) % in mild, moderate, severe and critical disease group (p-value 0.0001). Among 101 patients, 33(32.7%) required 1–5 L of oxygen while 25(24.8%) patients required more than 5 L of oxygen (p-value 0.0001). Radiological findings of pneumonia were present in 75(74.26%) patients, while 21(20.8%) patients had more than 50% lung infiltrate (p-value 0.0001).

The hematological and coagulation parameters in mild, moderate, severe and critical disease are shown in Table 2. Median(IQR) values of WBCs (p-value 0.004), ANC (p-value 0.002), NLR (p-value 0.001), D-dimer level (p-value 0.001), ferritin (0.0001), LDH (0.0001) were significantly increased in patients with critical disease as compared to the patients with mild, moderate and severe disease. Median (IQR) values of APTT (p-value 0.003) and CRP (p-value 0.0001) were suggestively increased in patients with severe disease.

Other parameters like Hb, MCV, HCT, ALC, Platelet count, prothrombin time did not show any significant association with severity of disease. (Table 2).

Mortality was observed in 8(7.9%) patients, out of them 6(5.9%) patients died.
patients were in critical disease while 2(%) were in severe disease (p-value 0.0001). All deaths were in severe and critical disease. Of the 101 patients, 7(6.9%) patients received mechanical ventilation. Out of 101 patients, 1(0.9%) patient was diagnosed as Thrombotic Thrombocytopenic Purpura and 1(0.9%) presented with deep venous thrombosis during admission. The median (IQR) values of statistically significant parameters WBC, NLR, D-Dimer level, ferritin and LDH were compared between patients with Oxygen saturation < 93% and patients with O2 saturation > 93%. The data was graphically presented and showed the suggestively increased levels of WBC, NLR, D-Dimer, CRP and ferritin in patients with Oxygen saturation level less than 93% Fig. 1.

4. Discussion

Results of the study illustrates hematological and hemostatic manifestations and their correlation with the severity of the disease in covid-19 patients. The study reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females. A study conducted by Jin JM et al., reported that males were more affected from the disease than females.

Our study demonstrated that Leukocytosis, neutrophilia and increased neutrophil to lymphocyte ratio, which might be due to inflammatory response, have a significant association with the disease severity. Neutrophil to lymphocyte ratio was highest in patients with critical disease. Liao D et al. also found elevated neutrophil to lymphocyte ratio as a useful predictor for severity and mortality of SARS-CoV-2 infection [10].

The association of NLR with severity of covid-19 disease was also concurred by a study of Yang AP et al., who concluded that high neutrophil to lymphocyte ratio and age are the independent factors for indicating poor clinical outcome of covid-19 patients [11].

We observed the association of APTT with worsening of disease and APTT was found prolonged in the severe and critical cases. Similar findings were observed by Iba T et al., evaluated that PT and APTT were either normal or deranged in the COVID-19 infected patients and these parameters depended upon the extent of coagulopathy as well as its association with other co-morbidities like Thrombotic Thrombocytopenic Purpura, Hemolytic Uremic Syndrome, Anti phospholipid Syndrome, Disseminated Intravascular Coagulation and Sepsis Induced Coagulopathy [14].

In our study, values of serum ferritin, LDH and CRP were significantly increased in severe and critical patients as compared to mild and moderate patient. In a retrospective cohort study from Wuhan, China, Terpo E et al., reported that increased ferritin and LDH were risk factors for Acute Respiratory distress syndrome, ICU support and mortality. Higher CRP has also been related to adverse aspects of COVID-19 disease, such as ARDS development, higher troponin-T levels and myocardial injury, and death [1,15].

Mortality rate in admitted patients was found in 7.9% patients, which is much more than the overall disease mortality in Pakistan. This mortality rate could be falsely high because it only protrudes the mortality among the admitted patients. Mehra MR et al., reported 5.8% death rate among the total of 8910 patients with covid-19 [16]. We observed in our study that most of the patients who expired had likely pulmonary embolism, but unfortunately, we could not investigate them.

One of the patients developed refractory thrombotic thrombocytopenic purpura who couldn’t survive even after 11 sessions of plasma exchange. Albion N et al., also documented autoimmune thrombotic thrombocytopenic purpura in a 57-year-old woman of covid-19 [17]. Hematological complications of the corona virus disease are associated with bad prognosis. However, we could not investigate the other hematologic

### Table 2

| Parameters                  | Units          | Mild Disease | Moderate Disease | Severe Disease | Critical Disease | p value |
|-----------------------------|----------------|--------------|------------------|----------------|------------------|---------|
| Hemoglobin (Hb)             | (g/dl)         | 13(1.9)      | 13(1.5)          | 13.05(2.4)     | 14.15(6.6)       | 0.648   |
| Mean Corpuscular Volume (MCV) | (FL)           | 85(5.3)      | 84(6.5)          | 84.3(6.8)      | 87.6(10)         | 0.293   |
| Hematocrit (HCT)            | (%)            | 40(5)        | 39.5(9)          | 38.6(7.3)      | 39.75(14.6)      | 0.39    |
| White blood count (WBC)     | (x 10^9/μl)    | 68.55(31)    | 83(15.38)        | 7.9(6.48)      | 14.99(6.46)      | 0.004   |
| Absolute neutrophil count (ANC) | (x 10^9/μl) | 6.12(5.02)   | 5.02(5.44)       | 6.44(5.87)     | 13.52(5.32)      | 0.002   |
| Absolute Lymphocyte count (ALC) | (x 10^9/μl) | 1.79(0.99)   | 1.48(1.09)       | 1.14(1.59)     | 0.91(1.44)       | 0.78    |
| Neutrophil to Lymphocyte ratio (NLR) | 3(5) | 7(4)         | 6.5(7)           | 11(21)         | 0.001            |         |
| Platelet Count              | (x 10^9/μl)    | 230(61)      | 245(104)         | 258(148)       | 226(220)         | 0.673   |
| Prothrombin time (PT)       | (seconds)      | 10.6(1.1)    | 10.71(0.9)       | 10.85(1.1)     | 11.4(2.2)        | 0.256   |
| Activated partial thromboplastin time (aPTT) | (seconds) | 25.3(3.3)    | 26.5(2.7)        | 27.9(5)        | 26.95(7)         | 0.003   |
| D-dimer                     | (μg/ml)        | 0.50(0.53)   | 0.64(0.92)       | 0.8(0.76)      | 1.88(1.08)       | 0.001   |
| Ferritin                    | (ng/ml)        | 150(151)     | 317(541)         | 584(779)       | 1436(1320)       | 0.0001  |
| Lactate Dehydrogenase (LDH) | (U/L)          | 170(111.5)   | 289(159)         | 463.5(487.5)   | 583.5(466.8)     | 0.0001  |
| C-reactive Protein (CRP)    | (mg/L)         | 4.40(20.85)  | 60(99.63)        | 99.5(205.1)    | 66.75(86.38)     | 0.0001  |
Fig. 1. Comparison of WBC, NLR, D-dimer, Ferritin and LDH between patients with O2 saturation more than 93% and patients with O2 saturation <93%. 

a) WBC

b) NLR ratio

c) D-Dimer level

d) Ferritin

e) LDH
parameters like fibrinogen, von Willebrand factor antigen and ADAM-TS 13 in our study because of low resources.

Limitation of the study was small sample size, because the covid-19 disease started to settle in Pakistan by end of July so we could not increase the number of patients. However, as the second wave of covid-19 is expected to hit the world, this study can provide a help in disease stratification in resource restraint countries.

5. Conclusion

The study concluded that Leukocytosis, neutrophilia, elevated Neutrophil to lymphocyte ratio, APTT, D-dimer, LDH and serum ferritin and CRP are significantly increased in patients with severe and critical disease. Hematological and coagulation manifestations are directly related to covid-19 disease and these markers may be utilized as useful prognosticator for early prediction of disease severity. Thus, appropriate management can be planned for such patients before the patient develops organ failure or shock.

Declaration of competing interest

The authors have no conflict of interest relevant to the manuscript.

Acknowledgement

We want to thank biostatistician Afshan Khanum for providing help in data analysis.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.amsu.2020.12.035.

Ethical approval

Ethical approval was taken from institutional Review Board. Certificate can be provided when required.

Funding and source of support

No funding was provided for the study.

Authorship Contribution

Sadia Taj: conceptualized and planned the study.
Ambreen Kashif: was involved in collection and organization of data.
Shehar Bano and Ayaz Lone: analyzed the data and helped in literature search.
Sadia Taj and Ambreen Kashif: prepared the manuscript.
Qasim Ahmed: reviewed and refined the manuscript.

Registration of research studies

1 Name of the registry:
2 Unique Identifying number or registration ID:
3 Hyperlink to your specific registration (must be publicly accessible and will be checked):

Guarantor

Dr. Sadia Taj.

Consent

Not applicable.

References

[1] E. Terpos, I. Ntanasis-Stathopoulos, I. Elalany, E. Kastritis, T.N. Sergentanis, M. Politou, et al., Hematological findings and complications of COVID-19, Am. J. Hematol. 95 (7) (2020) 834–847.
[2] P. Mehta, D.F. McAuley, M. Brown, E. Sanchez, R.S. Tattersall, J.J. Manson, et al., COVID-19: consider cytokine storm syndromes and immunosuppression, Lancet (London, England) 395 (10229) (2020) 1033.
[3] N. Tang, D. Li, X. Wang, Z. Sun, Abnormal coagulation parameters are associated with poor prognosis in patients with novel coronavirus pneumonia, J. Thromb. Haemostasis 18 (4) (2020) 844–847.
[4] Y. Shang, C. Pan, X. Yang, M. Zhong, X. Shang, Z. Wu, et al., Management of critically ill patients with COVID-19 in ICU: statement from front-line intensive care experts in Wuhan, China, Ann. Intensive Care 10 (1) (2020) 1–24.
[5] C. Huang, Y. Wang, X. Li, L. Ren, J. Zhao, Y. Hu, et al., Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China, The lancet 395 (10223) (2020) 497–506.
[6] D. Lillicrap, Disseminated intravascular coagulation in patients with 2019-nCoV pneumonia, J. Thromb. Haemostasis 18 (4) (2020) 786.
[7] G. Lippi, M. Plebani, R.M. Henry, Thrombocytopenia is associated with severe coronavirus disease 2019 (COVID-19) infections: a meta-analysis, Clin. Chim. Acta 506 (2020) 145–148, https://doi.org/10.1016/j.cca.2020.03.022.
[8] A. Angalal, M. Othman, Hemostatic laboratory derangements in COVID-19 with a focus on platelet count, Platelets (2020) 1–6.
[9] J.-M. Jin, P. Bai, W. He, F. Wu, X.-P. Liu, D.-M. Han, et al., Gender differences in patients with COVID-19: focus on severity and mortality, Front. Publ. Health 8 (2020) 152.
[10] D. Liao, F. Zhou, L. Liao, M. Xu, H. Wang, J. Xia, et al., Haematological characteristics and risk factors in the classification and prognosis evaluation of COVID-19: a retrospective cohort study, Lancet Haematol. 7 (9) (2020) e671–e678.
[11] A.-P. Yang, J. Liu, W. Tao, H-M Li, The diagnostic and predictive role of NLR, d-NLR and PLR in COVID-19 patients, Int. Immunopharmac. (2020) 106504.
[12] B.E. Fan, V.C.L. Chong, S.S.W. Chan, G.H. Lim, K.G.E. Lim, G.B. Tan, et al., Hematologic parameters in patients with COVID-19 infection, Am. J. Hematol. 95 (6) (2020) E131–E134.
[13] A. Bansal, A.D. Singh, V. Jain, M. Aggarwal, S. Gupta, R.P. Padapavil, et al., A Systematic Review and Meta-Analysis of D-Dimer Levels in Patients Hospitalized with Coronavirus Disease 2019 (COVID-19), medRxiv, 2020.
[14] T. Iba, J.H. Levy, M. Levi, J. Thachil, Coagulopathy in COVID-19, J. Thromb. Haemostasis 18 (9) (2020) 2103–2109.
[15] S. Imran, S.A. Fatima, N. Yunus, S. Taj, Association of plasma CRP level with the severity of COVID-19, Int. J. Sci. 9 (11) (2020) 20–23.
[16] M.R. Mehra, S.S. Desai, S. Kuy, T.D. Henry, A.N. Patel, Cardiovascular disease, drug therapy, and mortality in COVID-19, N. Engl. J. Med. 382 (2020) e102, https://doi.org/10.1056/NEJMoa200762.
[17] N. Albioli, R. Awol, R. Martino, Autoimmune thrombotic thrombocytopenic purpura (TTP) associated with COVID-19, Ann. Hematol. (2020) 1.