

Haematologic Profile Abnormalities and Coagulopathy Associated with Covid-19: A Prospective Study of 100 Patients

MNZ BHUIYAN^a, S GITI^b, MSM HOSSEN^c, MM RAHMAN^d, NE ZANNAT^e, S CHAKRABORTY^f

Abstract:

Background: The number of COVID-19 patients is exponentially increasing worldwide since December, 2019. Management in intensive care units (ICU) has become a major challenge; therefore, time demanding laboratory markers for early recognition of morbid forms is utmost important for timely triaging of patients.

Objective: The present study aimed to evaluate the role of haematological and Coagulation profile in the assessment of disease severity of COVID-19.

Methods: The current study is an observational study done prospectively in Armed Forces Institute of Pathology (AFIP) during the period of 28 March 2020 to 15 May 2020. The study subjects were haematological and coagulation profile data of 100 COVID-19 patients grouped in non-ICU and ICU at the baseline and at 4th day of admission in Combined

Military Hospital (CMH), Dhaka. All the data were recorded and analyzed by SPSS 20.0 programme.

Results: Here subjects with featured of lymphopenia were detected prominently in our COVID-19 ICU patients with a median nadir ALC of $0.9 \times 10^9/L$. The most common pattern of coagulopathy observed in patients hospitalized with COVID-19 is characterized by elevation in fibrinogen and D-dimer levels, particularly significantly higher in ICU group.

Conclusion: The present study is a reflection of significant anomalies in haematological and coagulation parameters during the disease process with COVID-19.

Key words: ALC-Absolute lymphocyte count, ANC- Absolute neutrophil count, Coronavirus, COVID-19, D-dimer, SARS-COV-2.

(J Bangladesh Coll Phys Surg 2020; 38: 61-66)
DOI: <https://doi.org/10.3329/jbcps.v38i0.47347>

Introduction:

The novel Coronavirus infection (now classified as COVID-19), first identified in December, 2019 in Wuhan, China, has contributed to significant mortality in several countries, with the number of infected cases increasing dramatically worldwide. The etiology for this outbreak is

a novel Coronavirus. It was isolated from human airway epithelial cells and named severe acute respiratory syndrome Coronavirus 2 (SARS-COV-2), which is responsible for the Corona Virus Disease 2019 (COVID-19).¹ The first COVID-19 case was detected in Bangladesh on 8th March, 2020. Most patients with COVID-19 predominantly have a respiratory tract infection, a proportion of patient progress to a more severe and systemic disease. Many patients with severe COVID-19 present with coagulation abnormalities that mimic other systemic coagulopathies associated with severe infections, such as DIC (Disseminated intravascular coagulation) or thrombotic microangiopathy, but COVID-19 has distinct features.² The main abnormalities in haematological parameters are lymphopenia.³ One of the most significant poor prognostic features in these patients is the development of coagulopathy. Organ insufficiency and coagulopathy were closely associated with high mortality.^{4,5} Up to now, clinical features and treatment modalities of patients infected with SARS-COV-2 have been reported meticulously. However, the relationship between SARS-COV-2 and laboratory parameters has been scarcely addressed. Present study is aimed to analyze the haematological and coagulation parameters and to evaluate the disease status of patients with COVID-19.

- Lieutenant Colonel Mohammed Nuruzzaman Bhuiyan, MBBS, MCPS, DCP (DU), FCPS (Haematology), Classified Specialist in Pathology, AFIP, Dhaka Cantonment.
- Major General Susane Giti, MBBS, MCPS, FCPS (Haematology), MMed (BSMMU), Classified Specialist in Pathology, Commandant, AFIP, Dhaka Cantt
- Lieutenant Colonel Mohammad Shameem Muntasir Hossen, MBBS, MCPS, DCP, FCPS (Haematology), Classified Specialist in Pathology, AFIP, Dhaka Cantonment.
- Lieutenant Colonel Md Moshir Rahman, MBBS, MCPS, DCP, FCPS (Haematology), Classified Specialist in Pathology, AFIP, Dhaka Cantonment.
- Major Nur-E-Zannat, MBBS, MCPS, DCP, AFMI, Dhaka Cantt.
- Major Sonia Chakraborty, MBBS, MCPS, AFMI, Dhaka Cantt.

Address of Correspondence: Lt Col Mohammed Nuruzzaman Bhuiyan, MBBS, MCPS, DCP (DU), FCPS (Haematology), Classified Specialist in Pathology, AFIP, Dhaka Cantonment. E-mail: dnruruzzaman4@gmail.com, Mob: 01769101147.

Methodology:

Study design: It is a prospective observational study.

Place of study: The study was carried out in Haematology department of Armed Forces Institute of Pathology (AFIP), Dhaka, Bangladesh.

Period of study: 28 March, 2020 to 15 May, 2020.

Study population: Between 28 March, 2020 to 15 May, 2020 around 635 patients with confirmed COVID-19 diagnosed at AFIP, were admitted to the CMH, Dhaka of which a total of 100 patients were included in the study.

Procedure of data collection: Haematological and coagulation profile data of 100 patients at the time of admission and 4th day of admission were collected by convenience sampling method and then data was analyzed and evaluated prospectively. We divided the patients into two groups- non-ICU group and ICU group (these patients were subsequently shifted from isolation ward to ICU). The laboratory findings were compared at baseline and 4th day of admission to assess the correlation between laboratory markers and disease severity. We obtained data from LIS (Laboratory information system) where we got information about age, gender and location of each patient.

Statistics: Data were processed using SPSS 20.0 statistical software. Non-normally distributed measurement data were presented using the median (interquartile range), and Fisher's exact t-test was used to compare between groups. Qualitative data were expressed as frequencies or rates, and the Fisher's exact test was used for comparisons between groups. Results with $P < 0.05$ were considered statistically significant.

Inclusion criteria:

1. Individuals with a positive COVID-19 by RT-PCR method aged > 20 years.
2. Subjects having available data regarding serial follow up of coagulation and haematological parameters.

Exclusion criteria:

1. Subjects having COVID-19 infection with short (2-3 days) hospital stay.
2. Patients with missing data regarding laboratory investigations, especially those who were minimally symptomatic.

Results:

Demographic Characteristics of patients with COVID-19

A total of 100 patients diagnosed with COVID-19 were included in this study. Among them 57 (57%) patients had fully recovered and were discharged from hospital. A number of 43 patients (43%) remained in hospital, of them 21 (21%) patients remained in isolation ward and 22 patients (22%) were shifted to ICU, subsequently 2 (2%) patients had died. These cases were divided into two groups based on requirements of ICU support; ICU group 22 patients (22%) including 2 death patients and non-ICU patients 78 (78%). There were 71 males (71%) and 29 females (29%). ICU and non-ICU groups showed statistically significant differences in sex ratio and age distribution. In the ICU group, there was more number of male 18 (81.8%). Male: female ratio in ICU patient were higher compared with the non-ICU patient (4.5:1 vs 2.12:1) (Table-I).

The median age was 34 years (Age range: 20-64 years). Notably ICU patients were about a decade older than the non-ICU patients. The median age of ICU patients was 50 years old while the median age of non ICU patients was 32 years old ($P=0.38$) (Table-II & table III).

Analysis of haematological parameters at admission

On admission, leucopenia was observed in 7 patients (7%). Lymphopenia featured in 13 patients (13%) with 10 patients having moderate lymphopenia (ALC $0.5-1 \times 10^9/L$) and 3 patients with severe lymphopenia (ALC $< 0.5 \times 10^9/L$). Those requiring ICU care had a lower ALC. Lymphopenia featured prominently in our COVID-19 ICU patient with a median nadir ALC of $0.9 \times 10^9/L$, compared to $1.8 \times 10^9/L$ in the non-ICU patient. Between the ICU (n-22) and non-ICU (n-78) patients using Fisher's exact tests, we found that admission ALC is discriminating laboratory indices with a P value of 0.006. The ICU patients in general presented with more profound lymphopenia with 9 out of 22 being lymphopenic but 2 of whom had severe lymphopenia. Notably, ICU patients during the hospitalization with a median peak ANC of $4.6 \times 10^9/L$ compared to $3.3 \times 10^9/L$ in the non-ICU patients (P value 0.039) (Table-IV).

The median nadir platelet count remained in the normal range ($> 150 \times 10^9/L$) for both groups of patients and was not a discriminating test on admission or during the hospital stay.

Characteristics of haematological parameters at 4th day after admission-

Comparing the haematological parameters at admission and at 4th day after admission in both ICU and non-ICU patients showed that median value of Hb, platelet, TLC and lymphocytes were decreased at 4th day in ICU patients while the median value of above parameters were constant or in some cases mildly decreased in non-ICU patients.

But the median value of neutrophil count was significantly increased (ANC of $11.6 \times 10^9/L$) at 4th day in ICU Patients while it was mildly increased non-ICU patients (Table-IV).

Analysis of abnormal coagulation parameters at admission

Coagulation profile of the 100 patients at admission revealed abnormal parameters in a total of 38 (38%) cases. The most common abnormal parameters were APTT (22/22%) followed by D-dimer (19/19%), FDP (15/15%), Fibrinogen (7/7%) and PT (5/5%). The proportion of abnormal coagulation parameters in the ICU patient was higher than in non-ICU patients (68.1% Vs 29.4%), of which proportion of abnormal APTT (40.9% vs 16.6%), D-dimer (40.9% vs 12.8%), FDP (22.7% vs 12.8%), Fibrinogen (13.6% vs 5.7%) and PT (13.6% vs 2.5%).

Comparison of coagulation function parameters at admission between the non-ICU and ICU patients showed that median value of FDP, Fibrinogen and D-dimer were higher in the ICU patients compared to the non-ICU patients. All differences were statistically significant, except PT and APTT (Table-V).

Characteristics of coagulation parameters 4th day after admission –

Comparing the coagulation and haematological parameters at admission and at 4th day after admission

showed that the median value of fibrinogen and FDP were increased both in ICU and non-ICU patients at 4th day but levels were significantly increased in ICU patients. The median values of D-dimer, PT, APTT were increased in ICU patients at 4th day, but those levels were constant or in some cases levels were improved in non-ICU patients (Table-V).

Notably the fibrinogen level >400 mg/dL were significantly higher in ICU patients than in non-ICU patients (13.6% vs 0%). The proportion of patients with elevated D-dimer <2 ULN (Upper limit of normal) was 76 (97.4%) in the non-ICU group but 18 (81.8%) in the ICU group. Further, in ICU group >22.7% of patients had elevated D-dimer >2 ULN, particularly the proportion with D-dimer >10 ULN was significantly higher in ICU groups than in non-ICU group (4.5% vs 0%).

Table-I

Distribution of sex among the study subjects (n=100)

Sex	Non-ICU	ICU
M	53	18
F	25	04
Ratio	2.12:7	4.5:1

Table-II

Distribution of age among the study subjects (n=100)

Age Range	ICU	Non-ICU
21-30	03	31
31-40	04	34
41-50	05	12
51-60	09	01
60+	01	00

Table-III

Demographic features of the study subjects

Parameters	Non-ICU patients (n=78)	ICU patients (n=22)	P-value (<0.05)	Overall (n=100)	
Age (Years)	Median (IQR)	Median (IQR)	0.038	34(29-42)	
	32(27-38)	50(35-55)			
Gender	M	53(67.9%)	18(81.8%)	0.013	71(71%)
	F	25(32.1%)	4(18.2%)	0.003	29(29%)

Table-IV

<i>Comparison of Haematological parameters between non-ICU group and ICU group</i>						
Haematological Parameters	Reference value	Non-ICU (n-78) Median (IQR)	ICU (n-22) Median (IQR)	P-value (<0.05)	Overall (n-100) Median (IQR)	
Admission	Hb (g/dL)	M:13-17 F:12-15	14.2(13.4-15.1)	12.8(11.2-13.7)	0.004	14(13-15)
	TLC(x10 ⁹ /L)	4-10	4.7(4-6.35)	5.8(5.25-11.45)	0.071	4.6(4.2-6.9)
	ALC(x10 ⁹ /L)	1-3	1.8(1.45-2.16)	0.9(0.5-1.24)	0.006	1.6(1.2-2.1)
	ANC(x10 ⁹ /L)	2-7	3.3(2.47-3.81)	4.6(3.72-12.1)	0.039	4.56(2.47-4.32)
	Platelets (x10 ⁹ /L)	150-410	189.5(151-226)	160(150-265)	0.019	181(151-223)
	Hb (g/dL)	M:13-17 F:12-15	11.95(11.1-14.25)	11.9(10.9-14.25)	0.038	12(11-14)
	TLC(x10 ⁹ /L)	4-10	6.1(5-7.6)	6.1(5-10.9)	0.253	6(5-8)
	ALC(x10 ⁹ /L)	1-3	1.6(0.8-1.8)	0.4(0.3-0.5)	0.027	1(0.8-1.5)
	ANC(x10 ⁹ /L)	2-7	4.2(2.8-4.8)	11.6(9.3-13.8)	0.012	4.5(2.9-6.1)
	Platelets (x10 ⁹ /L)	150-410	190.5(157-234)	191(157.5-264)	0.164	190.5(157-245)

Table-V

<i>Comparison of Coagulation Profile between non-ICU group and ICU group</i>						
Coagulation Parameters	Reference value	Non-ICU (n-78) Median (IQR)	ICU (n-22) Median (IQR)	P-value (<0.05)	Overall (n-100) Median (IQR)	
4 th day	PT (sec)	11-14	13.4(13-13.8)	13.8(13.4-14.8)	0.143	13.4(13-13.8)
	APTT (sec)	32-43	39.2(36.3-42.4)	41.2(40-48.7)	0.089	38.5(31.1-42.5)
	Fibrinogen (mg/dL)	200-400	279.5(240-316)	307.5(255-360)	0.042	280(240-323.5)
	FDP (µg/mL)	0-5.0	2.61(2.7-4.5)	3.28(2.85-5.4)	0.046	3.47(2.7-4.81)
	D-dimer(µg/mL)	0-0.5	0.29(0.2-0.39)	0.425(0.25-1.04)	0.038	0.31(0.2-0.47)
	PT (sec)	11-14	13(13-13.8)	13.4(13-14.8)	2.17	13.4(13-13.8)
	APTT (sec)	32-43	40.1(35.5-43.1)	42.5(40-46.7)	1.83	39.1(31.1-42.5)
	Fibrinogen (mg/dL)	200-400	280.5(270-290)	345(267.5-360)	0.016	290(267-350)
	FDP (µg/mL)	0-5.0	3.26(2.78-5.55)	4.8(2.57-4.8)	0.032	3.4(2.57-4.95)
	D-dimer(µg/mL)	0-0.5	0.72(0.34-0.47)	1.04(0.25-1.48)	0.029	0.32(0.27-1.04)

Discussion:

We analyzed the baseline data of 100 patients collected at the time of admission and at 4th day after admission. We divide the patient into two groups (ICU & non-ICU). The rate of requirement of ICU support in severe cases were 22% which was lower than previous reports.^{6,7} The ICU patients had a relatively higher proportion of male and were relatively older which is consistent with previous study.^{2,3,7} Data analysis of haematological parameters showed 13% of all patients presented with lymphopenia (ALC <1 x10⁹/L). This number is significantly smaller compared to 63% of patients in Wuhan, China, 28% of patients in Singapore, 42% of patients outside of Wuhan who presented with lymphopenia.^{6,8}

Those requiring ICU care had a lower ALC. These finding were also reported by Huang et al.⁶ on the characteristics of COVID-19 patients in Wuhan, China and also a study of haematological parameters in patients with COVID-19 in Singapore by Binguen Eugene Fan et al.⁹ Lymphopenia has been well described in retrospective analysis of patients in Hong Kong and Singapore affected with SARS-COV in 2003 and was associated with adverse outcome and ICU stay.^{10,11}

Analysis of coagulation profile showed that the incidence of abnormalities in conventional coagulation function parameters were higher in patients with ICU group compared to non-ICU group. The worsening parameters were observed at 4th day in ICU patients while the profile was constant or improving in non-ICU patients, suggesting that coagulopathy is more serious in ICU Patients and significantly correlate with the degree of disease severity to some extent.⁸ The incidence of abnormal fibrinogen level in 7% and the occurrence of abnormal D-dimer was 19% both in ICU and non-ICU patients. These results suggest that fibrinogen and D-dimer were not only increased in ICU patients but also increased to varying extend in non-ICU patients. However more significant increases were observed in patients with ICU patients. This finding is consistent with previous reports.^{8,12} Moreover we found that, level of fibrinogen, D-dimer and FDP level were increased at 4th day in ICU patients and non-ICU patients, but the level was significantly increased in ICU patients than non-ICU patients. The other parameters remained constant in non-ICU patients but all parameters were increased in ICU patients at 4th day

compared to admission. These indicators may be used as a biomarker for predicting disease outcome.

The limitation of our study is missing data as laboratory investigations were not performed daily on all patients especially those who were minimally symptomatic in the isolation ward. Correlating onset of symptoms with laboratory parameters is important which was not done in this study. There was no in-depth dynamic follow up and analysis of the relationship between coagulation function and prognosis in critically ill patients. Lastly, admission laboratory results for ICU patients transferred from other institutions were not reflected in our data set. Further multicenter study with a large sample size should be conducted comparing patients onset of symptoms and correlating their clinical condition to laboratory findings.

Conclusion:

COVID-19 is a systemic infection with a significant impact on the haematopoietic system and haemostasis. Careful evaluation of laboratory parameters at baseline and during the disease course can assist clinicians in formulating a tailored treatment approach and promptly provide intensive care to those who are in greater need. Larger analysis confirming these findings and investigating both the pathophysiology and impact of correction of coagulopathy on mortality are warranted.

References:

1. Zhu N, Zhang D, Wang W, Li X, Yang B, Sang S, et al. A novel coronavirus from patients with pneumonia in China, 2019. *New Engl J Med.* 2020; 382:727-733.
2. Tang N, Li D, Wang X, Sun Z. Abnormal Coagulation parameters are associated with poor prognosis in patients with novel Corona Virus pneumonia. *J Thromb Haemost.* 2020; 18:844-847.
3. Sohrabi C, Alsafi Z, O' Neill N, Khan M, Keruan A, Al-Jabir A, Losofidis C, et al. World health organization declares Global Emergency: a review of the 2019 Novel coronavirus (COVID-19). *Int J Surg.* 2020. Apr 2020; 76:71-76.
4. Chen N, Zhou M, Dong x, Qu J, Gong F, Han Y, et al. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. *Lancet.* 2020. March 2020; 395: 507-513
5. Guan Wj, Ni Zy, Hu Y, Ling Wh, Ou Cq, He Jx, et al. Clinical Characteristics of Coronavirus Disease 2019 in China. *N Engl J med* 2020; 382:1708-1720.
6. Hunag C, Wang Y, Li X, Chang Z, Xiong Y, Zhao Y, et al. Clinical features of patients infected with 2019 novel

- coronavirus in Wuhan, China. *Lancet*, 2020. Feb 2020; 395 (10223): 497-506.
7. Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, et al. Clinical Characteristics of 138 Hospitalized patients with 2019 Novel coronavirus-infected pneumonia in Wuhan, China. *JAMA*, 2020. March 2020; 323(11):1021-1104.
 8. Xu XW, Wu XX, Jiang XG, Xu KJ, Ying LJ, Mu CL, et al. Clinical findings in a group of patients infected with the 2019 Novel coronavirus (SARS-COV-2) out-side of Wuhan, China: retrospective case series. *BMJ* 2020; 368:600-606.
 9. Fan BE, Chang CL, Chan SSW, Lim GH, Lim KGE, Tan GB, et al. Haematologic parameters in patients with covid 19 infection at NCID, Singapore. *AMJ Haematol*. 2020; 95(6):1000-1002.
 10. Lee N, HUI DS, Wu A, Chan P, Cameron P, Joynt GM, et al. A Major outbreak of severe acute respiratory syndrome in Hong Kong. *N Engl J Med*. 2003; 348: 1986-1994
 11. Chng Wj, Lai HC, Earnest A, Kuperan P. Haematological parameters in severe acute respiratory syndrome. *Clin Lab Haematol*. 2005; 27: 15-20.
 12. Zhang JJ, Dong X, Cao YY, Yuan YD, Yang YB, Yan YQ, et al. Clinical characteristics of 140 patients infected with SARS-COV-2 in Wuhan, China. *Allergy* 2020; PMID:32077115.