

A Hospital-Based Assessment of the Hematological Parameters in Patients with Covid-19 Infection: An Observational StudyMd. Imteyaz Alam¹, Madhu Bharti², Manish Kumar Jha³, Poonam Kumari⁴¹Tutor, Department of Pathology, Darbhanga Medical College and Hospital, Darbhanga, Bihar, India²Tutor, Department of Pathology, Darbhanga Medical College and Hospital, Darbhanga, Bihar, India³Tutor, Department of Pathology, Darbhanga Medical College and Hospital, Darbhanga, Bihar, India⁴Associate Professor and HOD, Department of Pathology, Darbhanga Medical College and Hospital, Darbhanga, Bihar, India

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Conflict of interest: Nil

Abstract**Aim:** The aim of the present study was to evaluate the abnormalities in hematological parameters among severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infected patients in a tertiary care hospital.**Material & Methods:** This was a cross-sectional study carried out in Department of Pathology, The study period was of 12 months. Includes all patients confirmed with COVID-19 infection. A confirmed 100 case of COVID-19 was defined by a positive result on the reverse transcriptase polymerase chain reaction (RT-PCR) assay of a specimen collected on a nasopharyngeal swab. Informed consent was obtained from all patients during blood sample collection.**Results:** In the present study, a total of 100 patients confirmed with COVID-19 infection were included, out of which 60 (60%) were males and 40 (40%) were females. The majority of the patients (77%) were more than 40 years of age. In this study, all the patients including both males and females were categorized into three groups based on hemoglobin levels. Out of the 100 patients, 49 cases (49%) had adequate hemoglobin levels of above 12g/dl, around 43 cases (43%) had a mild reduction in the hemoglobin value, and only 8 cases (8%) had moderate anemia with hemoglobin levels less than 10g/dl. In this study, 68 cases (68%) had a WBC count in the normal range while 22 cases (22%) had leukopenia, and 10 cases (10%) had leucocytosis.**Conclusion:** The common hematological abnormalities in patients with COVID-19 infection were elevated NLR, lymphopenia, thrombocytopenia, and elevated D-dimer levels. Some of the hematological parameters including elevated NLR, thrombocytopenia, and lymphopenia have been found to correlate with the clinical severity of the SARS-COV-2 infection in many patients and are useful in monitoring the patients during the course of the disease.**Keywords:** c-reactive protein, schistocyte, prognosis, anemia, thrombocytopenia, neutrophil-lymphocyte ratio, lymphopenia, neutrophilia, leucocytosis, leukopeniaThis is an Open Access article that uses a funding model which does not charge readers or their institutions for access and distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>) and the Budapest Open Access Initiative (<http://www.budapestopenaccessinitiative.org/read>), which permit unrestricted use, distribution, and reproduction in any medium, provided original work is properly credited.**Introduction**

The novel corona viral infection resulting in severe acute respiratory syndrome (SARS) in humans has been declared a global pandemic by the World Health Organisation (WHO) and has affected more than one million people worldwide since its initial outbreak in Wuhan, China in December 2019. [1] SARS-CoV-2 is approximately 80% similar to SARS-CoV, invades host human cells by binding to the angiotensin-converting enzyme 2 (ACE2) receptor. [2] The disease has an incubation period of 2 to 5 days and spreads by droplet infection and surface contact. Electron microscopy and molecular diagnosis helped in better understanding of the pathology predominantly occurring in the

respiratory tract. [3] Endothelial inflammation, diffuse alveolar damage are associated with hyperreactivity and hyperinflammation of cellular immune system leading to severe hypoxia and acute respiratory distress syndrome. [4] While most patients develop an uncomplicated illness, approximately 1% to 4% develop severe lung involvement with a progressive reduction in oxygen saturation requiring admissions to the ICU. [5]

The ACE-2 receptors are also distributed in the heart, kidneys, liver, intestine, vascular endothelium, cerebral cortex, and hematopoietic cells. Hence SARS-CoV-2 infection will manifest

as a systemic disease with the involvement of multiple organ systems including gastrointestinal, neurological, immunological, cardiovascular, and hematopoietic systems. [6] Covid-19 has significant impact on pulmonary, cardiovascular, renal, gastro intestinal, neurological and on hematopoietic system. The impact of Covid-19 disease on Immune system and hemostasis has been studied. Recent preliminary data following the Covid-19 out-break indicated an association of complete blood count (CBC) parameters [7] and coagulation profile (increased D-Dimer, fibrinogen and (FDP) with disease progression. [8] A wide range of CBC abnormalities in COVID-19 have been observed and they include leukopenia or leukocytosis, thrombocytopenia, lymphopenia, neutrophilia with increased neutrophil-lymphocyte ratio (NLR), eosinopenia and abnormal platelet-lymphocyte ratio (PLR). [9] Peripheral smear examination revealed abnormal WBC morphology. Neutrophils showed abnormal nuclear segmentation, nuclear projections, fetus-like nuclei (COVID nuclei), and toxic granules. Monocytes showed cytoplasmic vacuoles.

Lymphopenia with occasional reactive large granular lymphocytes was seen in some cases. Thrombocytopenia with giant platelets was seen in some patients. The RBCs showed evidence of microangiopathic hemolysis in the form of fragmented RBCs, increased polychromasia, and nucleated RBCs in severe COVID-19 infection. [10,11]

Hence in this study, we examined various haematological parameters as prognostic markers to assess the disease progression, severity and outcome in COVID-19 patients.

Material & Methods

This was a cross-sectional study carried out in Department of Pathology, Darbhanga Medical College and Hospital, Darbhanga, Bihar, India. The study period was of 12 months. Includes all

patients confirmed with COVID-19 infection. A confirmed 100 case of COVID-19 was defined by a positive result on the reverse transcriptase polymerase chain reaction (RT-PCR) assay of a specimen collected on a nasopharyngeal swab. Informed consent was obtained from all patients during blood sample collection.

Inclusion Criteria

- Blood samples from individuals of all age groups and gender with a positive COVID-19 report by RT-PCR.

Exclusion Criteria

- Individuals with other causes of fever.

Methodology

All the blood samples received from patients with an RT-PCR-confirmed COVID-19 infection in the department of pathology during the two months study period were used for the study. This was a laboratory-based study where the hematological reports including CBC, NLR, serum ferritin, serum C-reactive protein (CRP), serum lactate dehydrogenase (LDH), and D-dimer levels of all the blood samples from COVID-19-positive patients received in the department of pathology were retrieved from the laboratory records. Three ml of blood was taken into ethylenediaminetetraacetic acid (EDTA) vacuettes for haematology investigations. EDTA samples were inverted several times to prevent coagulation. Haematological parameters were obtained using the 5 part haematology analyser (SYSMEX XT 2000i) The Leishman-stained peripheral smears of all those patients were reviewed. All the findings were tabulated and analyzed.

Statistical Analysis

Data were entered into Microsoft Excel (Microsoft Corp., Redmond, WA, USA) and analyzed using Statistical.

Results

Table 1: Age and gender distribution of patients

Age	Males (n)	Females (n)	Number of patients n (%)
≤ 20	4	0	4 (4%)
21 - 30	5	0	5 (5%)
31- 40	5	9	14 (14%)
41-50	11	9	20 (20%)
51- 60	10	10	20 (20%)
61- 70	15	8	23 (23%)
>71	10	4	14 (14%)
Total	60 (60%)	40 (40%)	100 (100%)

In the present study, a total of 100 patients confirmed with COVID-19 infection were included, out of which 60 (60%) were males and 40 (40%) were females. The majority of the patients (77%) were more than 40 years of age.

Table 2: Hemoglobin and WBC distribution among patients

Hemoglobin (g/dl)	Number of patients	Percentage
<10	8	8%
10 – 12	43	43%
>12	49	49%
Total WBC count (cells/ cu.mm)		
<4000	22	22%
4000 -11000	68	68%
>11000	10	10%

In this study, all the patients including both males and females were categorized into three groups based on hemoglobin levels. Out of the 100 patients, 49 cases (49%) had adequate hemoglobin levels of above 12g/dl, around 43 cases (43%) had a mild reduction in the hemoglobin value, and only

8 cases (8%) had moderate anemia with hemoglobin levels less than 10g/dl. In this study, 68 cases (68%) had a WBC count in the normal range while 22 cases (22%) had leukopenia, and 10 cases (10%) had leucocytosis.

Table 3: Distribution of NLR and platelet among patients

NLR	Number of patients	Percentage
< 2	10	10%
2 - 4	45	45%
5 - 7	25	25%
8 - 10	12	12%
>10	8	8%
Platelet count (cells/cu.mm)		
<1.5 lakhs	7	7%
1.5 - 4.0 lakhs	91	91%
>4.0 lakhs	2	2%

Out of 100 cases, 45 patients (45%) had high NLR values from 2-4 and 8 cases (8%) having markedly raised NLR of more than 10. In this study, the majority of cases (91%) had platelet count in the normal range while 7 cases (7%) had thrombocytopenia and 2 cases (2%) had thrombocytosis.

Table 4: Distribution of peripheral smear abnormalities among patients

S. No.	Peripheral smear features	Type of abnormalities	Number of cases (%)
1.	RBC abnormalities	Microcytic hypochromic anemia	6 (6%)
		Normocytic Normochromic anemia	10 (10%)
		Schistocytes	14 (14%)
		Leukocytosis	11 (11%)
2.	WBC abnormalities	Leukopenia	20 (20%)
		Neutrophilia	90 (90%)
		Lymphopenia	52 (52%)
		Lymphocytosis	1 (1%)
		Neutrophils with abnormal nuclear morphology (Ring nuclei)	82 (82%)
3.	Platelet abnormalities	Neutrophils with toxic granules	80 (80%)
		Thrombocytopenia	5 (5%)
		Thrombocytosis	2 (2%)
		Giant platelets	4 (4%)

The RBC abnormalities included normocytic normochromic anemia (10%), microcytic hypochromic anemia (6%), and the presence of schistocytes (14%). The WBCs showed various quantitative abnormalities with neutrophilia being the predominant abnormality seen in 90% of cases followed by lymphopenia in 52% of cases. The WBCs also showed morphological abnormalities in the form of neutrophils with ring nuclei (82%) and toxic granules (80%). The platelet abnormalities

included thrombocytopenia (5%), thrombocytosis (2%), and giant platelets (4%).

Discussion

Corona virus disease 2019 (COVID-19) is a systemic viral infection presenting mostly as acute respiratory illness with a high rate of hospitalization and mortality. The emergence and rapid spread of the deadly COVID-19 disease caused by severe acute respiratory syndrome coronavirus 2

(SARSCo2) is an evolving public health crisis worldwide. [12] Covid-19 has significant impact on pulmonary, cardiovascular, renal, gastro intestinal, neurological and on hematopoietic system. The impact of Covid-19 disease on Immune system and hemostasis has been studied. Recent preliminary data following the Covid-19 out-break indicated an association of complete blood count (CBC) parameters [13] and coagulation profile (increased D-Dimer, fibrinogen and (FDP) with disease progression. [14]

In the present study, a total of 100 patients confirmed with COVID-19 infection were included, out of which 60 (60%) were males and 40 (40%) were females. Patel et al [1] from India reported 70% of male patients, Bhuiyan et al [15] from Bangladesh reported 71% male patients while Araya et al [16] from Ethiopia reported 62.3% male patients. The majority of the patients (77%) were more than 40 years of age which was in concordance with the study by Patel et al. [1] Bhuiyan et al [15] reported the median age of patients admitted to ICU as 50 years and the median age of non-ICU patients as 32 years. In this study, all the patients including both males and females were categorized into three groups based on hemoglobin levels. Out of the 100 patients, 49 cases (49%) had adequate hemoglobin levels of above 12g/dl, around 43 cases (43%) had a mild reduction in the hemoglobin value, and only 8 cases (8%) had moderate anemia with hemoglobin levels less than 10g/dl. Patel et al [1] from India reported decreased hemoglobin in 36% of cases while Chen et al [17] from China reported a reduction in hemoglobin in 51% of cases which was almost similar to the present study. A meta-analysis by Lippi et al [18] revealed low hemoglobin concentrations among COVID-19 patients with severe disease than in mild infections. In this study, 68 cases (68%) had a WBC count in the normal range while 22 cases (22%) had leukopenia, and 10 cases (10%) had leucocytosis. In the study by Patel et al [1] from India, leukopenia was seen in 4% of cases while leucocytosis was seen in 20% of cases, unlike the present study. However, Wan et al [19] from China reported leukopenia in 20.7% of patients with COVID-19 infection similar to the present study.

Out of 100 cases, 45 patients (45%) had high NLR values from 2-4 and 8 cases (8%) having markedly raised NLR of more than 10. Elevated NLR signals the progression of pneumonia in COVID-19 patients and thus serves as an important prognostic parameter among COVID-19 patients. [20] In this study, NLR was elevated in 43% of cases out of which 6.1% cases had marked elevations in NLR of more than 10. Terra et al [21] from Brazil reported a higher death rate among COVID-19 patients with $NLR \geq 10$. In this study, the majority of cases

(91%) had platelet count in the normal range while 7 cases (7%) had thrombocytopenia and 2 cases (2%) had thrombocytosis. Zhou et al [22] from China reported thrombocytopenia in 7% of cases almost similar to the present study. There was a higher incidence of thrombocytopenia among the non-survivors than among survivors of COVID-19 infection in the study by Zhou et al. Various pathogenic mechanisms have been involved in the development of thrombocytopenia that includes a direct effect on platelet production by the cytokine storm associated with SARS-CoV-2 infection, increased consumption of platelets due to microthrombi formation in COVID-19 infection, or due to destruction of platelets by auto-antibodies. [23]

The RBC abnormalities included normocytic normochromic anemia (10%), microcytic hypochromic anemia (6%), and the presence of schistocytes (14%). The WBCs showed various quantitative abnormalities with neutrophilia being the predominant abnormality seen in 90% of cases followed by lymphopenia in 52% of cases. The WBCs also showed morphological abnormalities in the form of neutrophils with ring nuclei (82%) and toxic granules (80%). The platelet abnormalities included thrombocytopenia (5%), thrombocytosis (2%), and giant platelets (4%). In a study conducted by Pezeshki et al [24] from Iran, the most frequent peripheral smear abnormalities were thrombocytopenia (78.7%), smudge cells (67.4%), normocytic normochromic anemia (54%), giant platelets (42.7%), atypical lymphocytes (36%), leukopenia (37.1%), leukocytosis (29.2%) and schistocytes (27%).

Conclusion

The common hematological abnormalities in patients with COVID-19 infection were elevated NLR, lymphopenia, thrombocytopenia, and elevated D-dimer levels. Some of the hematological parameters including elevated NLR, thrombocytopenia, and lymphopenia have been found to correlate with the clinical severity of the SARS-COV-2 infection in many patients and are useful in monitoring the patients during the course of the disease. Awareness of the hematological abnormalities and their careful evaluation during the disease course will play a vital role in predicting the severity of the clinical illness and thereby guiding early intervention and appropriate intensive care to those who are in need. This will bring down the morbidity and mortality caused by SARS-CoV-2 infection to a considerable extent.

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