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"A STUDY OF CHANGES IN HEMATOLOGICAL PARAMETERS AND MORPHOLOGICAL ALTERATIONS IN LEUCOCYTES OF COVID-19 PATIENTS IN THIRD WAVE"

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ABSTRACT

Introduction: COVID-19 is a rapidly spreading pandemic caused by SARS-Cov-2. In the third week of November 2021, India experienced third wave. This study was taken up to show changes in Hematological parameters and morphological alterations in leucocytes in the third wave.

Material methods: This prospective observational study included confirmed cases of Covid-19, at Government medical college and hospital Miraj from 9-Jan-2022 to 13-Feb-2022 during third wave of Covid-19. Hematological parameters including Hb, TLC, DLC, Platelet count, NLR ratio and morphological changes in WBC were studied

Results: A total of 185 confirmed cases of Covid-19 patients for CBC were enrolled. 131 were males and 54 were females with median age of 70 years. Neutrophilia, lymphopenia, eosinopenia, raised ESR ratio with morphological alterations like hypolobated neutrophils, reactive lymphocytes having deep blue granular cytoplasm were reported.

Conclusion- Number of hospitalized cases and ICU admissions were less in the COVID-19 third-wave in India. Assessment of hematological parameters is important when investigating the COVID-19 patients as it will assist physicians in formulating the approaches to treatment and also essential in monitoring the disease progression.

Key words: COVID 19, lymphopenia, neutrophilia, eosinopenia

Introduction:

Coronavirus disease was first identified as a result of research in a group of patients who develop respiratory tract symptoms such as fever, cough, shortness of breath in Wuhan Province in China at the end of December 19.[1] On 02-Nov-2020 WHO named officially Virus

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as COVID-19. On 11th-March WHO declared COVID-19 as a pandemic disease. Since the middle of March 2021second wave has started and on April 09, the highest number of cases has been identified in India.[1,2] In the third week of November 2021, third wave started in India with around 7,000 to 9,000 daily new cases of Covid-19. In the last week of November, the emergence of Omicron which is the latest variant of SARS Cov2 was seen. Hospitalized cases were less in the COVID-19 third wave in India due to natural infections in the last two years, vaccination of maximum number of population and high transmissibility but less Virulence of the Omicron variant. COVID-19 symptoms might differ from person to person, resulting in a clinical presentation of disease ranging from asymptomatic to moderate infections, to major life-threatening instances needing ICU hospitalisation. Viral infections change morphology in number of cells in peripheral blood smear. The aim of present study is to investigate changes in hematological parameters and morphological alterations in WBC in COVID-19 positive patients during 3rd wave.

Materials and Methods:

The present study is conducted in Government Medical college and Hospital, Miraj which was declared as dedicated Covid-19 hospital on 29 March 2020. First case of Covid-19 was diagnosed on 25 March 2020 in Government Medical college and Hospital, Miraj. The present study is prospective study which included confirmed cases of Covid-19, diagnosed on nasopharyngeal swab by RT-PCR admitted at Government medical college and hospital Miraj from 9-Jan-2022 to 13-Feb-2022 during 3rd wave of Covid 19. We received samples in K2-EDTA anticoagulated tubes were processed on automated Sysmex XN-550 hematology analyzer. Peripheral blood films were prepared and stained with Leishman stain. Hematological parameters including Hb, TLC, DLC, Platelet count, NLR ratio were studied. WBC morphology changes were analyzed on peripheral blood smear examination.

Results:

A total of 185 confirmed cases of COVID-19 patients were included. Out of 185 cases131 were males and 54 were females. Male predominance is seen. In present study maximum number of cases were from age group 60-80 and least number of cases were from less than 20 yrs age group (Graph 1).

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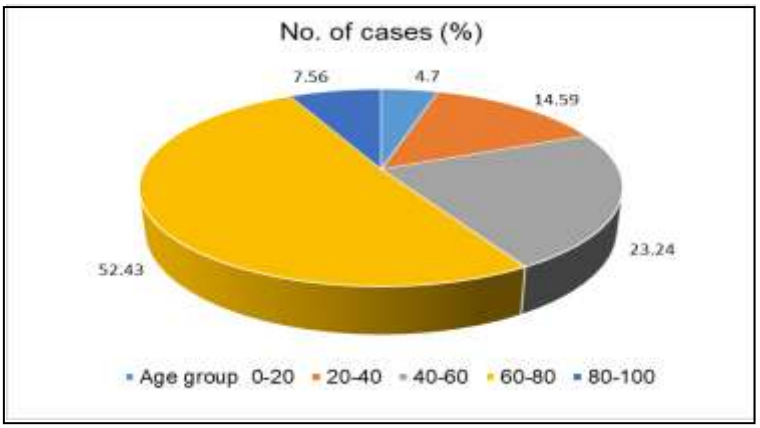
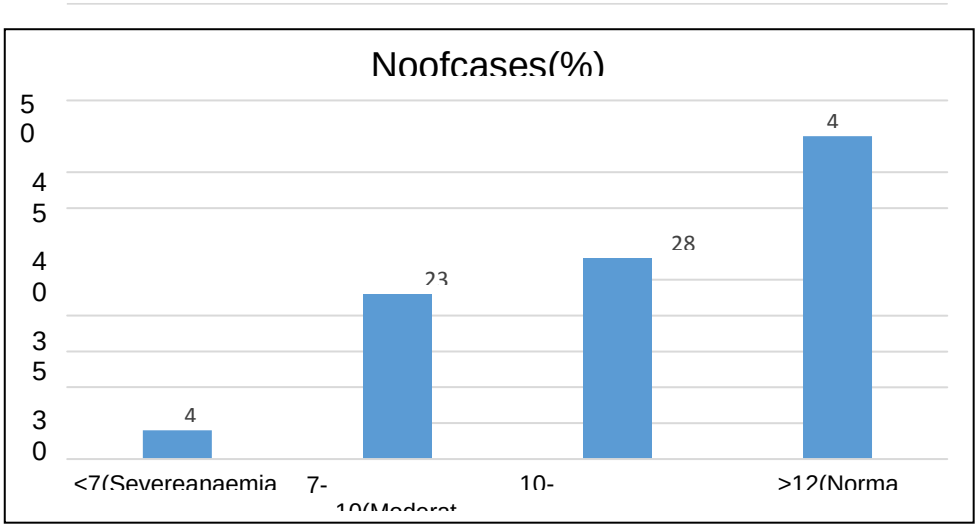
Graph 1: Age distribution of cases in present study



Patients were categorized patients into different groups based on Hemoglobin value, Hb value Less than 7gm/dl as severe anaemia, Hb value between 7gm/dl-10gm/dl as moderate anaemia, Hb value between 10gm/dl to 12gm/dl as moderate anaemia and Hb value more than 12gm/dl as normal. In present study maximum number of patients (45%) have normal Hb value, followed by patients with mild anaemia (28%), followed by patients with moderate anaemia (23%). Only 7 cases (3.7%) have severe anaemia (Graph 2).

Graph 2: Distribution of cases according to Haemoglobin in present study-



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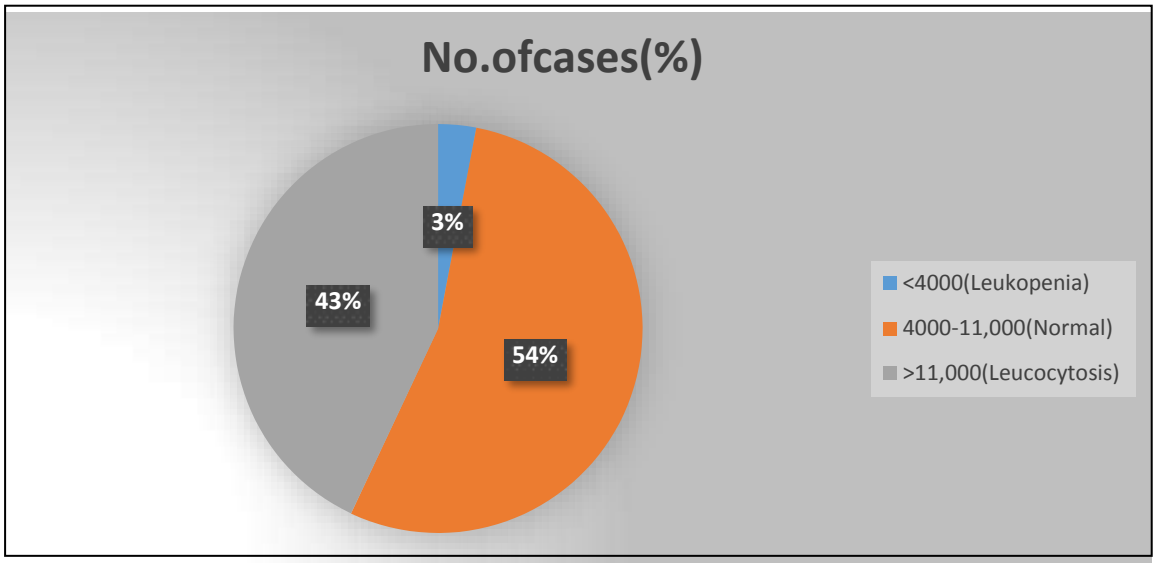
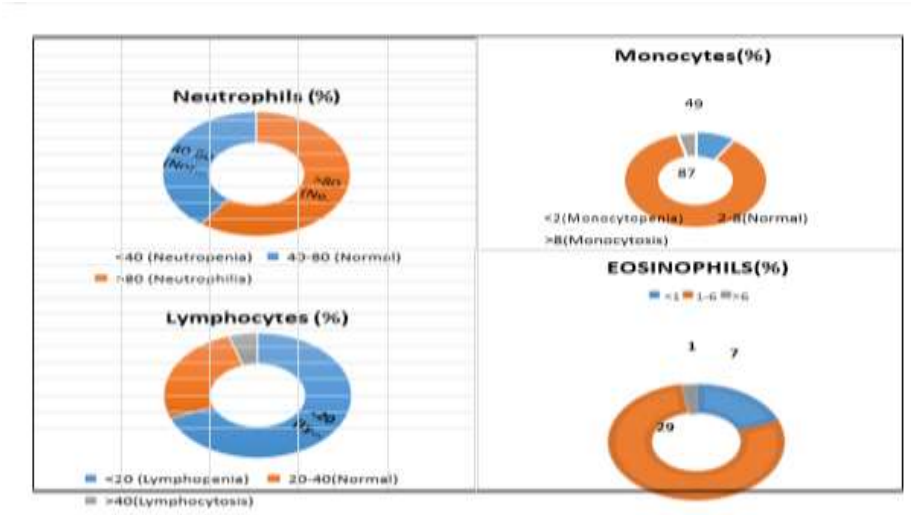
Maximum number of cases (54%) showed normal leucocyte count and 43% of cases had leucopoenia (Graph 3).



Graph 3: Distribution of cases of Total Leucocyte count in present study-

The maximum number of cases (60%) had neutrophilia and 40% cases had normal neutrophil count. In present study maximum number of cases (69%) had lymphopenia, 26% cases had normal lymphocyte count and 5% cases had lymphocytosis. 87% cases had normal monocyte count, 9% had monocytopenia and4% cases had monocytosis. Maximum number of cases showed Eosinopenia (70%), 29% cases showed normal eosinophil count and 1% showed eosinophilia (Graph 4).

Graph 4: Distribution of cases according to percentage of WBCs in present study



The maximum number of cases (74%) had normal platelet count, 19%cases had

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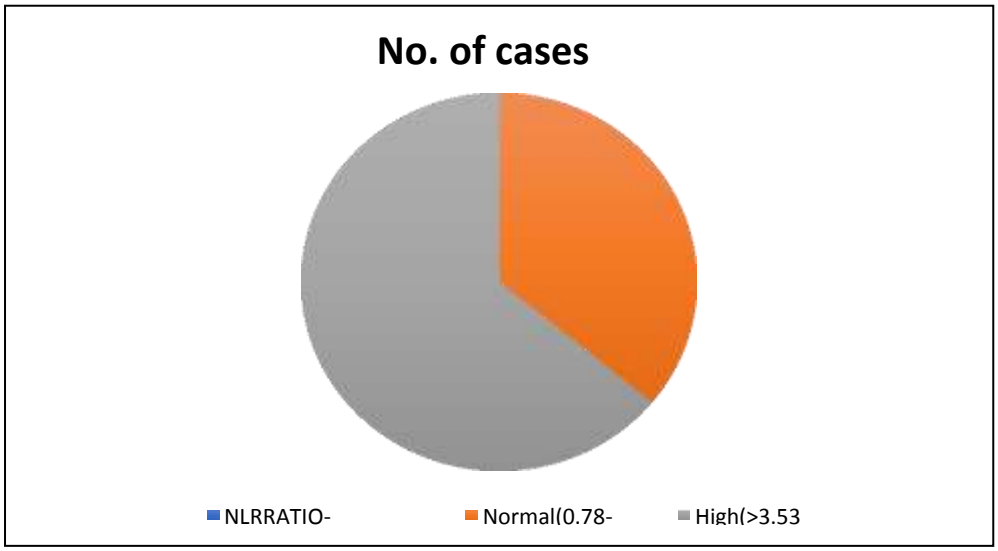
thrombocytopenia and 7% cases had thrombocytosis. In Neutrophil: Lymphocyes ratio maximum number of cases (64%) showed high NLR ratio and 36% cases showed normal NLR ratio. None of the patient had low NLR ratio in present study (Graph 5).

Graph 5: NLR ratio wise distribution of cases in present study



Peripheral smear examination is done in all COVID-19 positive patients. Various morphological abnormalities are noted in different leucocytes. Neutrophils in some cases showed hypolobated nuclei, C shaped nuclei with few of them were apoptotic. Few cases showed cytoplasmic coarse granules and vacuoles in neutrophils and monocytes. (Figure 1).

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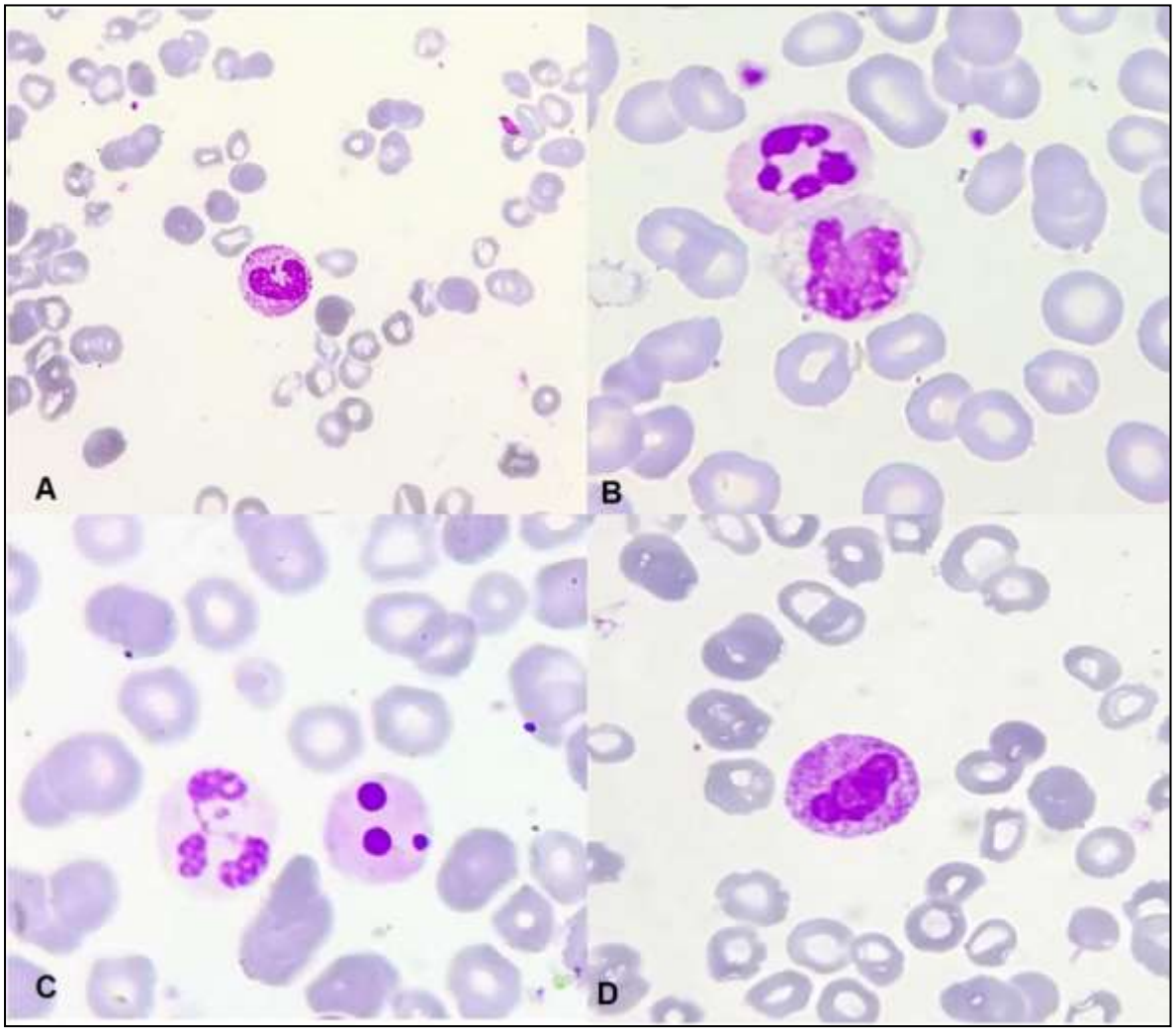
Figure1: Morphological Changes in Neutrophils



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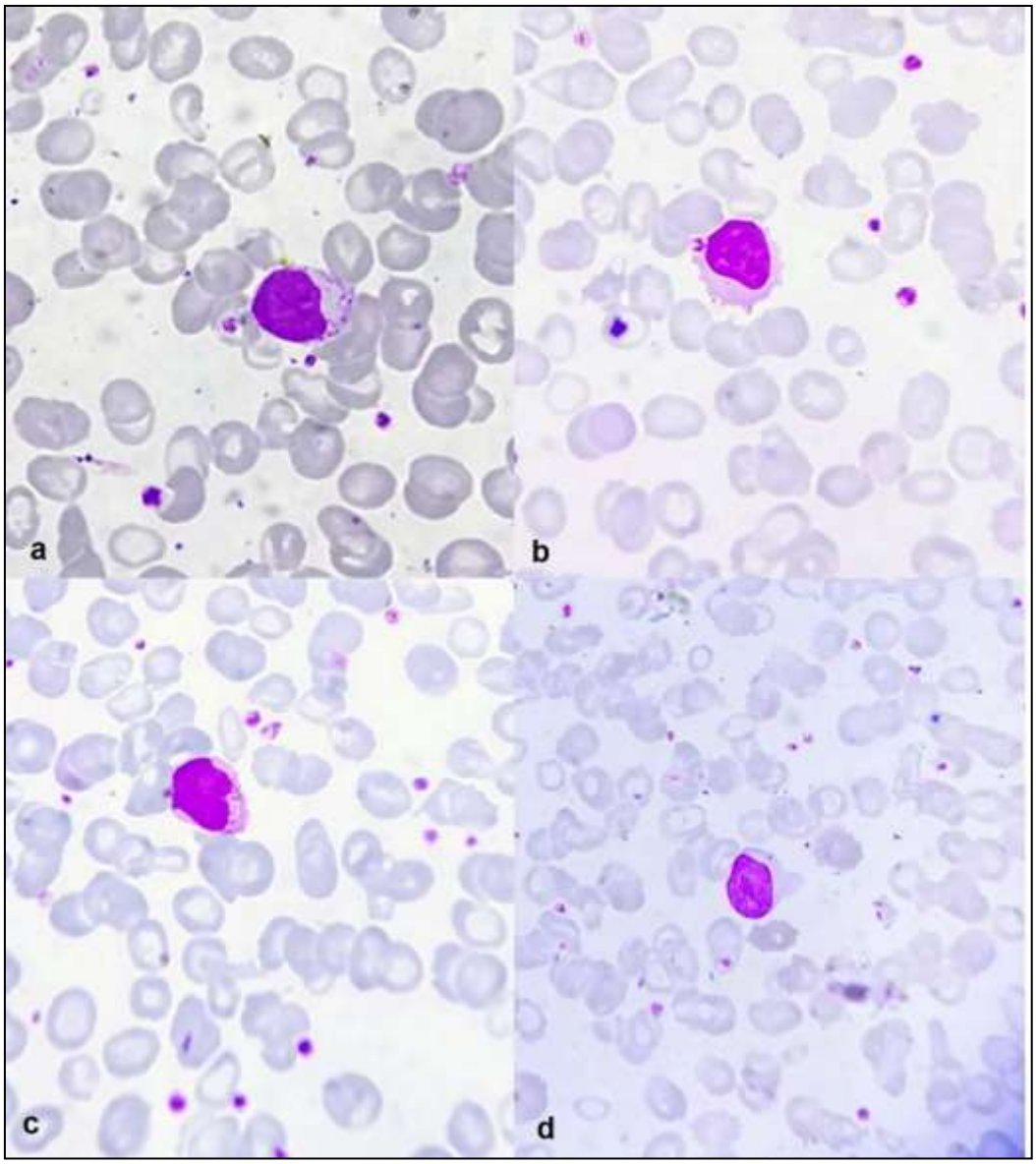
Reactive lymphocytes were noted in few cases. Some reactive lymphocytes showed abundant deep blue granular cytoplasm, indented nuclei. (Figure2).

Figure 2: Morphological Changes in Lymphocytes



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DISCUSSION:

Assessing clinical and hematological indicators is important when investigating the COVID 19 outbreak. Our study investigated hematological parameters such as Haemoglobin, WBC count, neutrophil count, monocyte count, eosinophils, platelets and NLR ratio. Of all 185 cases 70.81% are males and 29.19% are females, this resembles the studies done by E. Usul et al.,[3] and Qian K et al,[4] where males and females were 69.3% and 30.7%. Previous studies have also indicated that males are more susceptible to COVID 19 than females and have accounted for this in terms of lower expression of angiotensin converting enzyme – 2 receptors for corona virus in females.[5]

The maximum number of cases of COVID 19 patients in our study ranges between 60 -80 yrs. in 97 cases (52.43%). This means that COVID 19 in third wave targeted individuals above 60 years than individuals below 50-year age. This data is comparable with studies done by Abozer Y et al.,[5] and Yuan T et al.[6] The hematological parameters were analyzed in 185 cases of COVID 19 patients. In this study, maximum number of patients (45%) have normal haemoglobin (>12gm%). In contrast the study done by Usul E et al.,[3] shows that the COVID 19 positive patients have high haemoglobin values compared to normal individuals while Xu Y et al.,[7] study data shows that COVID 19 positive patients have low haemoglobin values compared to normal individuals. Concerning iron dysmetabolism in COVID-19, Ehsani et al., highlighted similarity between the distant amino acid sequence of SARS-CoV-2 spike glycoprotein cytoplasmic tail and the hepcidin protein.[8]

Corona viruses recognise host receptors with their spike proteins, allowing them to change shape and enter the host cytoplasm; coronaviruses may break their spike polypeptides utilising host furins and proteases, enabling cell entrance.[9] They found hepcidin mimicry by the virus would take place through this complex mechanism.[7] Leukocytosis is usually a sign that the body is fighting an infection, and the same is true in COVID-19. In our study, 54% patients have Total leucocyte count within normal limit and about 43% patients have leukocytosis. COVID-19 patients who had a high WBC count is reported in study done by Zhu B et al.[10] In contrast low WBC count is seen in study done by F. A. Naoum et al.[11] The primary function of neutrophiles clearance of pathogens and debris through phagocytosis.[12,13]

The release of neutrophil-chemo attractive elements and the resulting recruitment of

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neutrophils are a global host response to viral infection.[13] Upon SARS-CoV-2 infection, elevated numbers of neutrophils have been observed in studies done by L. Borges et al.,[14] Sun et al.,[15] and Wang B et al.[16] The present study shows maximum number of patients 111 (60%) having neutrophilia which is comparable with above studies. Our study shows lymphopenia in 127 cases (69%) which is comparable with studies done by Zhu B et al.,[10] Guan WJ et al.,[17] and Ding X et al.[18] Regarding the coronavirus, the neutrophil-to-lymphocyte ratio (NLR), a well-known marker of infection and systemic inflammation, has evidenced an enhanced inflammatory response in COVID-19 patients. Since the ARDS is the primary cause of mortality in patients with COVID-19, the elevated NLR values suggest a poor prognosis in COVID-19 disease.

Asghar et al.,[19] found an NLR value of 5.48 as the potential markers for severity of disease. In our study, we observed that 119 patients (64%) show high NLR ratio that is>3.53 which is relatable to the studies done by C. Qin et al.,[20] Sun et al.,[15] Wang B et al.,[16] and B. J. Barnes et al.[21] Severe cases of COVID-19 appear to be related to increased NLR levels, whether NLR could be an independent predictor of mortality in COVID-19 patients requires further investigations.

In addition, present study includes distribution of cases according to percentage of monocytes and eosinophils. About 160 cases (87%) shows normal monocyte count and about 130 cases (70%) shows eosinopenia. In first and second wave many of the studies like Singh A et al.,[21] Terposet al.,[22] Fan BE et al.,[23] shows monocytopenia and eosinopenia. Platelets may also directly interact with SARS-CoV-2 and have been shown to carry the SARS-CoV-2 virus. Platelets can also facilitate the virus uptake by secretion of the subtilis in-like proprotein convertase furin. In this study 137 cases (74%) shows normal platelet count. Similar to our study, other studies reported that platelet values were found to be normal in many patients at the time of hospital admission.[24]

Some studies have found a relationship between thrombocytopenia and the severity of the COVID-19andrelated mortality. Various morphological abnormalities are noted in different leucocytes in present study. Neutrophils in some cases showed hypolobated nuclei, C shaped nuclei with few of them were apoptotic. Few cases showed cytoplasmic coarse granules and vacuoles in neutrophils. Reactive lymphocytes were noted in few cases. Some reactive lymphocytes showed abundant deep blue granular cytoplasm, indented nuclei. Similar morphological abnormalities in leucocytes are seen in Singh A et al.,[22] Ilhami B. et

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al.,[25] and Olga P et al.,[26]

Conclusion:

The third wave of COVID-19 was noted with less number of hospitalized cases and ICU admissions in India due to natural infections in the last two years, vaccination of maximum number of population and high transmissibility but less Virulence of the Omicron variant. Peripheral smear examination is rapid, inexpensive investigation to assess COVID-19 positive patients. Neutrophilia, lymphopenia, eosinopenia, raised NLR ratio and morphological alterations are seen in COVID-19 positive patients. Assessment of hematological parameters is important when investigating the COVID 19 outbreak.

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Conflict of Interest: Nil

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