

Haematological Parameters in Patients of Covid-19 from Baltistan

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ABSTRACT

Objective: To determine hematological parameters and C-reactive protein (CRP) in patients of Corona virus disease 2019 (COVID-19) from Baltistan.

Study Design: Cross-sectional study.

Place and Duration of Study: Combined Military Hospital, Skardu Pakistan, from Jul to Sep 2020.

Methodology: One hundred and ninety-four patients presenting with signs and symptoms suggestive of COVID-19 were included. They were divided into two equal groups on result of polymerase chain reaction (PCR) for severe acute respiratory syndrome corona virus 2 (SARS-CoV-2). PCR positive patients were included in Group-A and PCR negative in Group-B. Blood samples of patients from both groups were tested for total leukocyte count (TLC), absolute neutrophil count (ANC), absolute lymphocyte count (ALC), neutrophil to lymphocyte ratio (NLR), haemoglobin (Hb), platelet count, prothrombin time (PT), activated partial thromboplastin time (APTT), D-Dimer and CRP. Results of both groups were compared using appropriate statistical tests.

Results: Statistically significant difference was found between mean values of TLC, ANC, NLC and platelet count of both groups. Mean±SD value of TLC, ANC, NLR and platelet count of groups A and B were $6.91\pm 2.26 \times 10^9/l$ and $8.90\pm 3.79 \times 10^9/l$, $4.71\pm 2.20 \times 10^9/l$ and $6.77\pm 3.68 \times 10^9/l$, 3.99 ± 4.38 and 5.73 ± 5.46 , $196.52\pm 64.71 \times 10^9/l$ and $229.09\pm 132.43 \times 10^9/l$, respectively. The difference in mean values of ALC, Hb, PT, APTT, D-Dimer and CRP of both groups was not statistically significant.

Conclusion: In patients with symptoms suggestive of COVID-19, total leukocyte count, absolute neutrophil count, neutrophil to lymphocyte ratio and platelet count may help in diagnosis.

Keywords: Absolute Neutrophil Count, COVID-19, C-Reactive Protein, Neutrophil to Lymphocyte Ratio, Polymerase Chain Reaction.

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INTRODUCTION

The World Health Organization (WHO) declared COVID-19 as a pandemic in March 2020.¹ Worldwide, total number of cases reported till 30 January 2021 is about 101.5 millions and deaths caused by COVID-19 are about 2.1 million. In Pakistan, first confirmed case was reported in February 2020 and total number of reported COVID-19 cases is 544 thousand till 30 January 2021 with 11.6 thousand (2.13%) deaths.^{2,3}

Body aches, fever and cough are common symptoms.⁴ Patients may also experience symptoms like flu, headache, tiredness, loss of taste and smell, loose motions, conjunctivitis and skin rash.^{5,6} Old age, asthma, chronic obstructive pulmonary disease, hypertension, diabetes, chronic renal failure and immunosuppressive states increase the risk of mortality and morbidity.⁶ The most common complication of COVID-19 is pneumonia. Other

complications include acute respiratory distress syndrome, acute kidney injury, myocarditis, superadded infections and multi-organ failure.⁷ The definitive diagnosis of COVID-19 is by PCR. Mortality in COVID-19 patients increases with high levels of CRP, D-dimers, interleukin-6 and ANC.⁸ Treatment is mostly supportive and only FDA approved drug is the antiviral drug remdesivir. Emphasis has been laid mostly on preventive measures.⁹ WHO has recommended wearing of a face mask, social distancing, avoiding unnecessary gatherings, early detection and isolation of cases and contacts, frequent hand washing and practicing droplets precaution.¹⁰

Complete blood count (CBC) and CRP are low-cost tests and available almost everywhere. If biomarkers of CBC and CRP help in diagnosis of COVID-19, they may be used as initial screening test in patients with symptoms suggestive of COVID-19. Keeping in view, present study was conducted to determine hematological parameters and CRP in COVID-19 patients.

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METHODOLOGY

This cross-sectional study was carried out at Combined Military Hospital Skardu, Pakistan, from Jul to Sep 2020. The study was conducted after taking approval from Ethical Review Committee of the hospital (ERC ref no 01/2021).

Inclusion Criteria: Patients of either gender presenting with signs and symptoms suggestive of COVID-19 were included.

Exclusion Criteria: Outpatient department (OPD) patients and paediatric age group (<12 years of age) were excluded.

Patients were recruited using non-probability consecutive sampling, after taking informed consent. WHO calculator was used to calculate sample size, which came to 194. They were divided into two equal groups on basis of result of PCR of NP and/or OP swab for SARS-CoV-2. SARS-COV-2 RNA was extracted manually and PCR was carried out on Sacycler using Sansure amplification kits. Positivity of PCR samples was decided as per guidelines of the kit manufacturer. Those patients who tested positive for SARS-COV-2 PCR were included in Group-A and those who tested negative were placed in Group-B.

Blood samples of both groups were tested for hematological and inflammatory markers. Total leukocyte count (TLC), hemoglobin (Hb), platelet count, Absolute Neutrophil Count (ANC) and absolute lymphocyte count (ALC) were estimated using Sysmex XP-100 haematology analyzer. Neutrophil to Lymphocyte Ratio (NLR) was calculated by dividing ANC with ALC. Prothrombin time (PT), Activated partial thromboplastin time (APTT), D-Dimer and C-reactive protein (CRP) were carried out manually as per standard protocols. D-Dimer and CRP was reported positive if value was more than 200 ng/ml and 6 mg/l respectively.

Data was analyzed using Statistical Program for Social Sciences (SPSS) version 20. For qualitative variables like gender, CRP and D-Dimer, frequency and percentages were calculated. For quantitative variable like age, ANC, ALC and Hb etc. mean and standard deviation was calculated. Chi square test was used to association categorical variables of both groups like CRP and D-Dimer. For numerical variables, independent sample t test was used to compare results of both groups. A *p*-value ≤0.05 was considered significant.

RESULTS

Of 194 patients, 167(86%) were male and 27(14%) were female. In Group-A, 88(90.7%) patients were male and 9(9.3%) were female (Figure-1). In Group-B, 79(81.4%) were male and 18(18.6%) were female (Figure-2). Mean age of Group-A was 41.94±18.85 years and that of Group-B was 42.68±16.88 years (*p*=0.275). Mean±SD value of TLC, ANC, NLR and platelet count of groups A and B were 6.91±2.26 × 10⁹/l and 8.90±3.79 × 10⁹/l, 4.71±2.20 × 10⁹/l and 6.77±3.68 × 10⁹/l, 3.99±4.38 and 5.73±5.46, 196.52±64.71 × 10⁹/l and 229.09±132.43 × 10⁹/l, respectively. Mean values of TLC, ANC and platelet count of Group-A patients were significantly lower than those in Group-B whereas mean value of NLR was significantly higher in Group-A (Table-I).

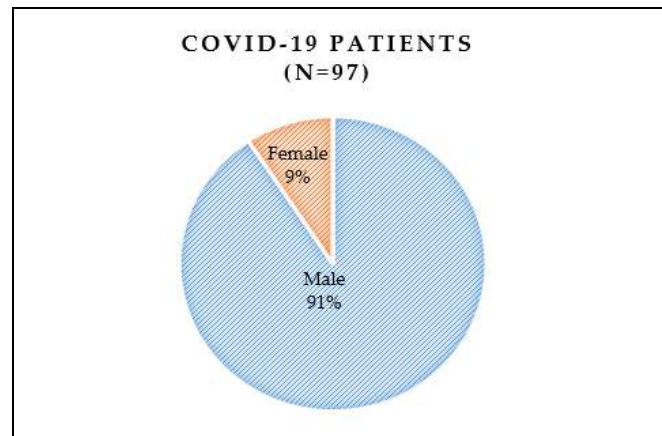


Figure-1: Gender distribution among Patients of COVID-19

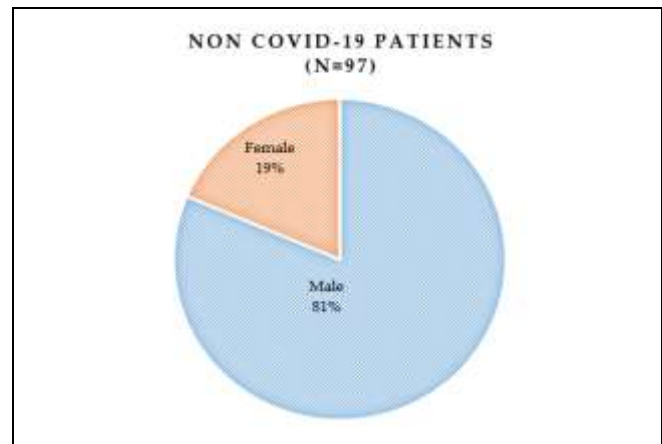


Figure 2: Gender distribution among those without COVID-19

Mean value of ALC, Hb, PT and APTT of groups A and B were 1.71±0.83 × 10⁹/l and 1.65±0.91 × 10⁹/l, 14.75±1.80 g/dl and 14.50±1.94 g/dl, 14.90±1.28 sec and 14.80±1.44 sec and 34.90±1.38 sec and 34.50±1.20

sec, respectively. There was no statistically significant difference found between mean values of ALC, Hb, PT and APTT of both groups (Table-I).

Table-I: Comparison of Quantitative Variables across Groups (n=194)

	Polymerase Chain Reaction Result	Mean±SD	p-value
Age (years)	Positive	41.94±18.85	0.275
	Negative	42.68±16.88	
TLC (x 10 ⁹ /l)	Positive	6.91±6.90	0.0136*
	Negative	8.90±3.78	
ANC (x 10 ⁹ /l)	Positive	4.71±2.19	<0.0001*
	Negative	6.77±3.68	
ALC (x 10 ⁹ /l)	Positive	1.71±0.83	0.953
	Negative	1.65±0.91	
NLR	Positive	3.99±4.38	0.015*
	Negative	5.73±5.46	
Hb (g/dl)	Positive	14.75±1.80	0.389
	Negative	14.50±1.94	
Platelet count (x 10 ⁹ /l)	Positive	196.52±64.71	0.016*
	Negative	229.09±132.43	
PT (sec)	Positive	14.90±1.28	0.686
	Negative	14.80±1.44	
APTT (sec)	Positive	34.90±1.38	0.084
	Negative	34.50±1.20	

*p-value < 0.05

TLC (total leukocyte count); ANC (absolute neutrophil count); ALC (absolute lymphocyte count); NLR (neutrophil lymphocyte ratio); Hb (haemoglobin); PT (prothrombin time); APTT (activated partial thromboplastin time)

In Group-A, CRP was elevated in 35 patients and normal in 62 patients whereas CRP was elevated in 34 patients and normal in 63 patients in Group-B ($p=0.574$) (Table-II). D-Dimer was elevated in 10 patients of Group-A and 8 patients of Group-B whereas 87 and 89 patients in groups A and B respectively had normal D-Dimer values ($p=0.621$) (Table-III).

Table-II: Association of C Reactive Protein positivity across Groups (n=194)

	Group-A n(%)	Group-B n(%)	p-value
CRP Positive	35(36.1%)	34(35.0%)	0.574
CRP Negative	62(63.9%)	63(65.0%)	

*CRP (C-reactive protein), PCR (Polymerase chain reaction)

Table-III: Association of D-Dimer positivity in Group A and B (n=194)

	Group-A n(%)	Group-B n(%)	p-value
D-Dimer Positive	10(10.3%)	8(8.2%)	0.621
D-Dimer Negative	87(89.7%)	89(91.8%)	

DISCUSSION

In the COVID-19 pandemic, a number of patients reported to our hospital with sign and symptoms suggestive of the COVID-19. However, it was almost impossible to segregate them into COVID-19 or non-COVID-19 patients solely on the basis of clinical

findings. With the availability of thermal cycler and equipment required for PCR at our set up, it became easy to separate the two groups. We carried out pertaining laboratory investigations of these patients as well and compared results of both groups.

The results of our study are in concordance with a meta-analysis comparing laboratory parameters in COVID-19 and non COVID-19 patients, which showed significantly lower TLC, ANC and platelet count in COVID-19 patients and no difference in ALC, D-Dimer and CRP among both groups.¹¹ Hengeveld *et al.*, reported significant difference between mean values of TLC, ANC and platelet count of COVID-19 and non COVID-19 patients, but no significant difference was found between mean values of ALC in both groups. Mean values of TLC, ANC and platelet count were lower in COVID-19 patients when compared to non-COVID-19 patients.¹² These findings are comparable to our study, except for mean value of ALC which was higher in Group-A as compared to Group-B. Similarly, Mardani *et al.*, observed a significantly lower mean value of TLC and higher neutrophil percentage in COVID-19 patients ($n=70$).¹³ Nalbant *et al.*, found that mean values of NLR and CRP were significantly higher and ALC was significantly lower in COVID-19 patients when compared with non-COVID-19 patients. No significant difference was observed between mean values of TLC, ANC and platelet count of both groups.¹⁴ In contrast, TLC, ANC, NLR and platelet count were significantly lower in COVID-19 patients as compared to non-COVID-19 patients in our study. Tan *et al.*, observed that mean values of TLC, ANC and NLR were lower in COVID-19 patients as compared to patients with influenza A and B.¹⁵

On study reported that mean value of platelet count was significantly lower in COVID-19 patients ($234 \times 10^9/l$) than non-COVID-19 patients ($250 \times 10^9/l$); similar to findings in our study.¹⁶ Same observations were reported by Soraya *et al.*¹¹

Patients of COVID-19 have elevated level of inflammatory biomarkers like CRP, IL-6, ferritin and procalcitonin. In our study, however, we tested for CRP because it was the only inflammatory marker test available at our set up. Different mechanisms have been proposed for high level of CRP in COVID-19 patients.¹⁷ Its production is increased by liver in the early phase of disease and comes down to normal in the recovery phase. The level of CRP rises with the disease severity and high mortality has been observed in patients with raised CRP level. COVID-19 patients

with raised CRP suffered from disease complications more frequently. Elevated level of CRP has also been linked with lung tissue destruction and increased need for oxygenation and ventilation.¹⁸ Likewise, many studies have reported elevated CRP level in patients of COVID-19 and its positive correlation with disease severity.^{13,15,18} In our study, CRP was positive in 36% of COVID-19 patients but no difference was found between COVID-19 and non-COVID-19 group, which is consistent with another study.¹¹

Coagulation abnormalities observed in COVID-19 patients include elevated D-dimer and fibrinogen levels with mild derangements of PT and APTT.¹⁷ Elevated D-Dimer values at admission may predict bleeding or thrombotic complications.¹⁸ Nalbant *et al.*, observed no significant difference in PT, APTT and D-Dimer values of COVID-19 and non-COVID-19 patients, which is consistent with results of our study.¹⁴

LIMITATIONS OF STUDY

The main limitations of our study are that the PCR positive group included patients of severe as well as non-severe disease and we did not study the relationship of hematological and inflammatory parameters with the disease severity and mortality. Out of the inflammatory markers, we studied only CRP because it was the only test available in our set up.

CONCLUSION

In patients with symptoms suggestive of COVID-19, total leukocyte count, absolute neutrophil count, neutrophil to lymphocyte ratio and platelet count may help in diagnosis.

Conflict of Interest: None.

Funding Source: None.

Authors' Contribution

Following authors have made substantial contributions to the manuscript as under:

JZC & SAS: Data acquisition, data analysis, critical review, approval of the final version to be published.

SAKK & FNS: Study design, data interpretation, drafting the manuscript, critical review, approval of the final version to be published.

Authors agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

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