COMPARISON OF INFLAMMATORY MARKERS WITH DIFFERENT LEVELS OF SEVERITY OF COVID-19 DISEASE

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ABSTRACT

Objective: To assess correlation of various inflammatory markers with disease progression in COVID-19.
Study Design: Cross sectional study.
Place and Duration of Study: The study was conducted at Pak Emirates Military Hospital (PEMH), Rawalpindi, from Apr 2020 to May 2020.
Methodology: All the patients admitted during this period were recruited in the study. Samples were collected from patients in medical wards, high dependency unit (HDU) and intensive care units (ICU). Sample was collected in plain tube for analysis of serum ferritin, quantitative C-reactive protein (CRP), and procalcitonin. For analysis of plasma lactate, sample was collected in sodium fluoride tube. Comparison of inflammatory markers amongst various groups was done to assess the association and correlation of these markers with the progression of disease.
Results: A total of 127 cases were studied, 114 were males and 13 were females. Age of patients in mild group was 41.6 ± 13.2, in moderate 53.3 ± 15.4 and in severe cases it was 54.6 ± 14.4. C-reactive protein and ferritin levels were significantly deranged in all groups with *p*-value of <0.001 respectively and same was the case for lactate and procalcitonin. Post Hoc analysis of the significant parameters showed that levels of all the parameters were significantly associated with all the stages of disease.
Conclusion: Majority of the Patients with COVID-19 disease exhibited elevated levels of inflammatory markers and their values significantly increased as the disease progressed with the time. As increase in inflammatory markers correlate with disease severity, regular monitoring by using these parameters can improve the disease outcome.

Keywords: C-reactive protein, Inflammatory markers, Lactate, Interleukin-6, Procalcitonin.

INTRODUCTION

The novel corona virus disease is spreading rapidly and causing havoc and consuming the health resources[1]. It is affecting both upper and lower respiratory tracts[1]. It spreads the same way as other corona viruses do by contact from one person to the other, but it is more contagious[1]. This virus causes infections which can be categorized as mild, moderate, and severe. Other forms of diseases by similar category of the viruses are the sudden acute respiratory syndrome and Middle East respiratory syndrome[4]. The common cold is also due to one of the types of the corona viruses[5].

Recent studies show that inflammatory response is playing a major role in the progression of the disease[6]. Inflammatory responses result into the recruitment of the monocytes and macrophages that cause the induction of the cytokines release[7]. These cytokines and chemokines are involved in the activation of the immune response that leads to cytokine storm which destroy tissues and cause organ failure[8]. These inflammatory markers have the potential to serve as the biomarker for monitoring and assessing the disease progression[9]. However, these results need further evaluation in monitoring the treatment plan and progression of the disease[10].

Rationale of this study was to identify the specific inflammatory markers which are raised

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and can indicate the severity of the disease. Our study assessed the association between the elevated levels of inflammatory markers and the progression of the disease and help the physicians in monitoring and evaluating the progression of the disease.

METHODOLOGY

This cross sectional study was conducted at Pak-Emirates Military Hospital, Rawalpindi. Data collection was done for the 127 patients, over a period of 5 weeks from, 15th April 2020 to 20th May 2020, having Real time-PCR positive for COVID-19 admitted in the hospital over a period of one month consecutive sampling was done for data collection. Patients were categorized as mild moderate or severe according to the symptoms as well as the location of the patient in different wards. Patients in regular wards were considered as mild, while those in HDU were considered to have moderate disease. Whereas patients admitted to ICU/ITC were considered as cases with Severe disease. Clinical data was obtained from wards and 3cc of venous blood sample was drawn in Vacctue TM Gel tubes by trained staff nurse under aseptic measures and were sealed and sent to lab. The samples were analyzed for C-reactive protein, lactate and procalcitonin using Roche Cobas 6000 based on spectrophotometric technique. Serum ferritin levels were assayed on Roche Cobas e411 by electrochemiluminescence. To ensure quality of results, quality control material of 2 different levels were used. All these patients were also followed for the outcome.

All the data was obtained after approval of ethical review board (ERC/ID/32). Informed written consent was taken from all patients. Their identity was not disclosed to anyone as each patient was allocated a specific identification number which was entered in data instead of patients' names.

Mean and SD were calculated for numeric data while frequencies and percentages were calculated for qualitative data. ANOVA was applied to test the association of parameters under observation with disease severity; Post Hoc Tukey's test was applied to test the intragroup association with various test perimeters. Pearson's correlation was applied to assess the relation of inflammatory markers with disease progression.

RESULTS

A total of 127 patients were included in the study, of which 114 (89.7%) were males and 13 (10.3%) were females (table-I). Mean age of the study participants was reported to be 41.6 ± 13.2 years (table-II). Our results showed that younger population had mild disease as compared to older people. All the parameters were significantly increased with the progression of the disease specially C-reactive protein and ferritin levels (table-II).

Post Hoc analysis of the significant parameters showed that levels of all the parameters were significantly associated with all the stages of disease (table-III). Outcome of disease was categorized based on patients discharged, expired and under treatment after a period of one month

Table-I: Gender distribution amongst different groups of disease.

Gender	Mild	Moderate	Severe
Male	27 (21.2%)	58 (45.6%)	29 (22.8%)
Female	5 (3.9%)	1 (0.07%)	7 (5.5%)

Table-II: Association of age and inflammatory markers with disease severity.

	Mild	Moderate	Severe	*p*-value
Age	41.6 ± 13.2	53.3 ± 15.4	54.6 ± 14.4	<0.001
C- Reactive protein (mg/L)	30.0 ± 64.3	74.5 ± 81.1	133.8 ± 102.2	<0.001
Ferritin (ng/ml)	312 ± 271.88	1164 ± 1879.9	2070 ± 1631.9	<0.001
Lactate (mmol/L)	1.52 ± 0.75	2.50 ± 1.28	3.53 ± 2.57	<0.001
Procalcitonin (ng/mL)	0.169 ± 0.068	0.564 ± 0.298	1.514 ± 1.59	<0.001

from the date of admission. The mortality reported in our patients was 0.8% (table-IV). Pearson's correlation showed that there is a significant and positive correlation of all the markers with severity of the disease, using ferritin as a marker for progression of disease.

Table-III: Post hoc analysis of significant parameters.

Inflammatory marker	Comparison groups		*p*-value
C- Reactive Protein	Severe	Moderate	0.003
		Mild	<0.001
	Mild	Moderate	0.045
Ferritin	Severe	Moderate	<0.001
		Mild	<0.001
	Mild	Moderate	0.034
Lactate	Severe	Moderate	<0.001
		Mild	<0.001
	Mild	Moderate	0.032
Procalcitonin	Severe	Moderate	0.016
		Mild	<0.001
	Mild	Moderate	0.034

Table-IV: Disease outcome in different stages of diseases.

	Mild	Moderate	Severe
Recovered & discharged	21 (16.5%)	59 (46.5%)	5 (3.9%)
Still under treatment	11 (8.7%)	-	29 (22.8%)
Death	-	-	1 (0.8%)

Table-V: Pearson's correlation of inflammatory markers with disease severity.

Marker	Correlation	*p*-value
C- Reactive protein	Pearson Correlation	0.400
	p-value	<0.001
Lactate	Pearson Correlation	0.457
	p-value	<0.001
Procalcitonin	Pearson Correlation	0.370
	p-value	<0.001

DISCUSSION

COVID-19 disease, caused by SARS-CoV-2, is rapidly spreading worldwide[1]. Despite the fact that most of the cases have mild clinical features and a good prognosis, It can result in acute respiratory syndrome and even death. To date no effective therapy for COVID-19 disease has been proposed[11,12]. Therefore, it is important to highlight the markers monitoring the progression of disease thus categorizing the patients and devising management protocols accordingly[6,7]. Several studies have shown increased levels of pro-inflammatory cytokines in serum of COVID-19 patients[13,14]. Also, anti-inflammatory agents for COVID-19 therapy highlight the critical role of inflammation in the progression of COVID-19[15]. However, the role of inflammatory markers in monitoring the severity of COVID-19 is still controversial[16].

As we know that C-reactive protein levels are increased significantly in acute inflammation, infection, and tissue damage, it can be used as indicator of inflammation. In our study C-reactive protein levels were significantly elevated as the disease severity progressed which is in concordance with previous studies[10,17,18]. Our study showed significant raise in levels of procalcitonin with increasing severity. Similar findings were reported in previous studies[13-15,18].

Although all the inflammatory markers showed a marked increase in levels with severity of the disease, but lactate levels showed maximum raise in critical cases as the Pearson's correlation suggests that it has the highest correlation coefficient. Rise in these levels was due to cytokine storm in patients with severe disease as suggested by Aloisio *et al*[19]. A meta-analysis by Gong *et al*, suggested a significant increase in inflammatory markers in critical patients. They emphasized the importance of C- Reactive protein, procalcitonin, ferritin and lactate apart from IL-6. They concluded that the above markers were also significantly elevated as the disease progressed[1].

It has been reported that there is cytokine storm in this infection. Cytokine storm so far is reported only in conditions where there is an impressively powerful activation of immune response. Uptill now, the clinical implications of the storm are less known[20].

Zeng *et al*, analyzed a total of 16 studies and evaluated more than 3000 patients with COVID pneumonia. Their results showed elevated levels

for C-reactive protein, procalcitonin, ESR and serum ferritin in critical patients as compared to those in mild group[1]. Higher viral load is also found in the cases reported having stronger cytokine and chemokine response attributing to the factor that these immune mediators are directly related to severity of the disease, similarly, higher mortality is also reported in cases with cytokine storm[21], inferring that vigilant monitoring of inflammatory marker is required in moderate to sever and particularly in critical patients. Keeping in view the results and limitations of our study, further study can be planned with larger sample size to avoid population bias in different study groups.

CONCLUSION

Majority of the patients with COVID-19 disease exhibited elevated levels of inflammatory markers and their values significantly increased as the disease progressed with the time. As increase in inflammatory markers correlate with disease severity, regular monitoring by using these parameters can improve the disease outcome.

CONFLICT OF INTEREST

This study has no conflict of interest to be declared by any author.

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