

Association of Mortality and Raised Inflammatory Markers such as Serum LDH, Serum Ferritin and CRP in Hospitalized Patient with Covid-19 Infection

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ABSTRACT

Objective: The objective of the study was to analyze correlation of serum ferritin, CRP (c-reactive protein) and lactate dehydrogenase with mortality in hospital due to infection caused by covid-19. Also, the aim was to evaluate the best cut off values.

Study Design: It was a cross-sectional study carried out in the medicine department of Civil Hospital and Khairpur Medical College, Khairpur. The study was done between June to September 2020.

Methodology: Patients that were admitted due to SARS COV-2 which was positive after PCR testing were the part of this study. Patients that had suggestive findings on radiograph but negative PCR test for SARS COV-2, patients with incomplete data or patients that left hospital against medical advice were not included in this study. At the time of admission, serum levels of ferritin, CRP and LDH were tested. The viral load of SARS COV-2 was analyzed through nasopharyngeal swab. WHO guidelines were used to evaluate the disease severity.

Results: In this study, 248 participants were taken with mean age 42.28± 17.84 years. 163 patients (65.7%) showed up with mild symptoms, 38 patients (15.32%) revealed moderate while 47 patients (18.95%) were severe cases. 27 patients (10.8%) expired in the hospital. The serum levels of ferritin (124 patients; 50%), CRP (86 patients; 34.67%) and LDH (179 patients; 72.17%) were elevated which was more frequent in patients with moderate or severe conditions and mortality. The 95% confidence interval and the areas in curves associated with receiver operating characteristics for serum levels of ferritin, CRP and LDH were 0.920 (0.840-0.98), 0.91 (0.84-0.95) and 0.87 (0.75-0.92) respectively. CRP (≥45.5 mg/L) had 87.46% sensitivity and 89.10% specificity; ferritin (≥723 ng/ml) had 94.22% sensitivity and 87.60% specificity and the LDH (≥428.5 U/L) had 91.50% sensitivity and 81.65% specificity to predict the mortality.

Conclusion: The levels of inflammatory markers at the time of admission will help predict mortality in Covid-19 patients.

Key Words: Inflammation, Coronavirus, Outcome, mortality, Pakistan

INTRODUCTION

This pandemic of covid-19 has become the emergency globally. The clinical spectrum of the disease varies from asymptomatic condition to the failure of respiratory system and requiring support from ventilator. Mortality rate is the main concern of the disease^{1,2}. According to WHO, (6 oct 2020) the confirmed cases of covid-19 were 35,274,993 and mortality rate was 1,038,534 in about 216 countries^{3,4}. The mortality around the globe was 2.9% and in Pakistan it was 2.06% according to the gov. of Pakistan. Inflammation is the most important factor in the disease pathogenesis and its progression. The inflammatory cytokines as well as chemokines were released by macrophages which further cause infiltration of monocytes and the neutrophils inside the lungs^{5,6}. This damages the alveolar cells and the endothelium which further leads to hypoxia and finally acute respiratory distress syndrome^{7,8}. CRP inflammatory marker was found to be usually elevated in the disease as a response for protection. The complement pathway is activated when this marker binds to the phosphocholine on the surface of damaged cell, causing the virus and the damaged cells to be eradicated

from the host. The causes of mortality in the disease include the cytokine storm defined as the increased levels of various cytokines like IL-6 or interferon γ . IL-6 is the main cytokine involving in mortality, but Pakistan lacks setups to measure it routinely⁹⁻¹⁰. CRP is the mainstay factor in diagnosing the cytokine release syndrome. Inflammation can worsen the disease to an extent that the patient can either need ventilatory support or may expire within three days. The mortality rate can be decreased by frequent follow ups of the covid patients and keen monitoring of the clinical presentation to approach effective treatment in severe cases¹¹. So, lab and clinical evaluation are important diagnostic factors.

Many studies observed the prognostic evaluation of various inflammatory markers in the disease. IL-6 and procalcitonin are difficult to be interpreted due to its less availability or unfeasibility, especially in Pakistan. The serum levels of CRP, ferritin as well as LDH can be monitored in all labs easily. The results obtained may be of great help in identifying the correct approach of intervention and follow ups¹². Since, covid-19 is a new type of infection, therefore, the studies lack any local data to evaluate correlation between the inflammatory markers and clinical

spectrum as well as outcome in various severe cases. The aim of this study was to analyze association between serum levels of ferritin, CRP and LDH and mortality in hospital admitted patients and to evaluate the best cut off values to predict mortality.

METHODOLOGY

It was a cross-sectional study carried out in the medicine department of Civil Hospital and Khairpur Medical College, Khairpur. In this study, 248 participants with mean age 42.28± 17.84 years were included. The study was approved by Ethics Review Committee of the Hospital. The patient’s consent was not required as the obtained data lack potential to reveal identification of any patient. The SARS COV-2 was diagnosed on the basis of positive results from PCR testing. The probable cases explained as the CT findings suggestive of covid-19 with negative PCR results for SARS COV-2, patients with incomplete data or patients that left hospital against medical advice were not included in this study.

Evaluation was done qualitatively based on its presence or absence. In the first half hour of hospital admission, blood was drawn from patients to analyze through tests including CRP, Ferritin and LDH. Data was collected from labs as well as hospitals medical records. Missing information was searched through Laboratory information management system software. As shown in Table-I, the disease severity was evaluated as per WHO guidelines. Except few patients that presented with critical condition, the group was merged into severe diseased group.

The data was reviewed through IBM SPSS statistics for windows of version twenty (IBM Corp, Armonk, NY). The qualitative data was presented as percentages as well as numbers, and the quantitative data as mean values of standard deviation. Continuous variable having

nonparametric distribution were analyzed by median and interquartile ranges. Shapiro-Wilk test was used to evaluate normality of the obtained data. The median and the interquartile ranges of inflammatory markers in mild, moderate, or severe diseased patients were compared through Kruskal-Wallis test. Independent-samples Mann-Whitney U-test was done to analyze the inflammatory marker levels among the groups that were made on the basis of outcome. Chi-square/ Fisher’s exact test was done to compare the group of patients having increased levels of inflammatory markers. The ROC (receiver operating characteristic) curve evaluation was done to estimate the sensitivity as well as specificity of markers of inflammation to estimate mortality and to recognize the optimal values of cut off points.

Curves were formed and IBM SPSS statistical analysis was carried out to determine AUC of inflammatory markers. This is quite good in determining differentiation between various classes such as a high AUC for inflammatory marker in this study means good distinction between death of people in hospital or people discharging from hospital. The KaplanMeier method evaluate the discharge probability of hospitalized patients as various inflammatory markers predict at the same thresholds. The p value was taken as less than 0.05 which was considered as significant. The online study dataset is available in Harvard dataverse.10

RESULTS

In this study, 248 participants were taken with mean age 42.28± 17.84 years. 163 patients (65.7%) showed up with mild symptoms, 38 patients (15.32%) revealed moderate while 47 patients (18.95%) were severe cases. 27 patients (10.8%) expired in the hospital.

Clinical severity of COVID-19 infection given in Table-I

Category	Definition
Mild	Symptomatic patients fulfilling case definition for COVID-19 without evidence of pneumonia or hypoxia.
Moderate	Clinical signs of pneumonia (fever, cough, dyspnea) but no signs of severe pneumonia, including SpO2 ≥90% on room air.
Severe	Clinical signs of pneumonia (fever, cough, dyspnea) plus one of the following: respiratory rate >30 breaths/min; severe respiratory distress; or SpO2 <90% on room air.
Critical	Acute respiratory distress syndrome.

Table II shows Clinical characteristics.

Variable	Value	
Age (years)	42.28± 17.84	
Gender	Male	213 (85.89%)
	Female	35 (14.11%)
Disease severity at admission	Mild	163 (65.73%)
	Moderate	38 (15.32%)
	Severe/critical	47 (18.95%)
Duration of hospital stay (days)	12.16± 6.80	
Mortality*	Total	25 (10.08%)
	Mild disease	1 (4.00%)
	Moderate disease	3 (12.00%)
	Severe/critical disease	21 (84.00%)

*p-values <0.05 for comparisons between the three groups.

The serum levels of ferritin (124 patients; 50%), CRP (86 patients; 34.67%) and LDH (179 patients; 72.17%) were elevated which was more frequent in patients with moderate or severe conditions and mortality.

Table III shows the Comparison of inflammatory markers amongst different groups

	All patients (n=248)	Outcomes			Clinical disease severity			
		Discharged (n=221)	Death (n=27)	P	Mild (n=163)	Moderate (n=38)	Severe (n=47)	p
CRP (mg/l)	6.25 (2.28 – 29.23)	5.6 (2.13-22.40)	131.5 (75.85-190.30)	<0.001	4.00 (1.65 – 8.60)	29.5 (6.20-54.35)	103 (28.40- 156.60)	<0.001
Ferritin (ng/ml)	239 (128.00- 613.85)	227 (119.00-449.35)	1768.6 (1218.65- 2198.60)	<0.001	183 .00 (99.20- 288.60)	481.6 (271.85- 847.60)	1312 (621.60- 1925.20)	<0.001
LDH (U/l)	331 (237.6- 445.6)	312 (235.00-412.35)	659 (543.60-814.60)	<0.001	275 (231- 362)	418 (283.85- 610.60)	501 (359.60- 722.60)	<0.001

All values represent median and interquartile range.

Table-III shows inflammatory marker values. The 95% confidence interval and the areas in curves associated with receiver operating characteristics for serum levels of ferritin, CRP and LDH were 0.920 (0.840-0.98), 0.91 (0.84-0.95) and 0.87 (0.75-0.92) respectively. CRP (≥ 45.5 mg/L) had 87.46% sensitivity and 89.10% specificity; ferritin (≥ 723 ng/ml) had 94.22% sensitivity and 87.60% specificity and the LDH (≥ 428.5 U/L) had 91.50% sensitivity and 81.65% specificity to predict the mortality.

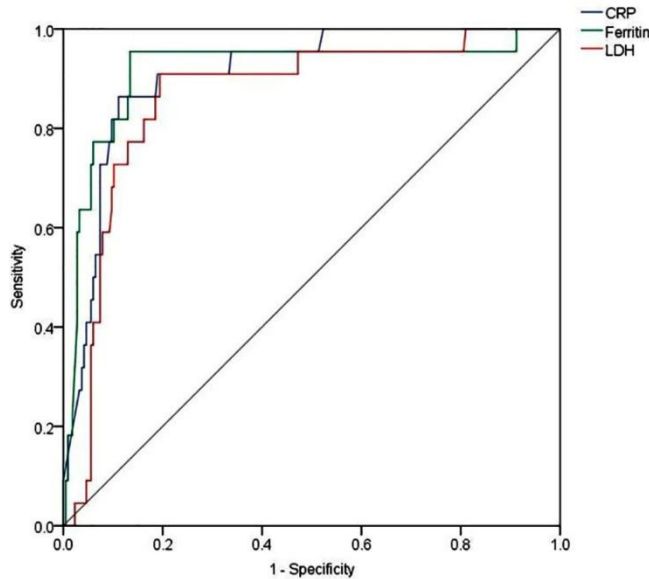
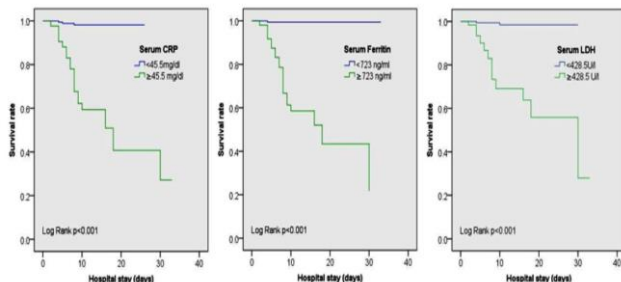


Figure 1: ROC curves for different inflammatory markers

Fig 2 shows Kaplan–Meier survival analysis on discharge probability of hospitalized patients as various inflammatory markers predict at the same thresholds.



DISCUSSION

The results displayed the elevated inflammatory markers in moderate or severe diseased patients in comparison with patients that had mild infection. Keeping cytokine storm in mind as pathogenesis of covid-19, it was also analyzed and concluded that there was no significant difference between moderate or severe form in association with inflammatory markers¹³.

In contrast to other studies, the severe disease patients had higher level of inflammatory markers than moderate forms¹⁴. this may be because of categorization based on upper limit of severity spectrum. However, severe and moderate diseased patients need treatment at hospital while mild diseased form can be managed at homes¹⁵.

In a study of USA, the moderate and severe form of disease does not reveal much difference in CRP levels. Ferritin levels were found to be least elevated in patients with moderate or severe diseased form. But ferritin level helps more in predicting the mortality with high AUC as well as odd ratio. Elevated level of serum ferritin was related to the bad outcomes in this study¹⁶⁻¹⁷. The high ferritin level in covid infection may be due to increased production due to cytokines such as tumor necrosis factor- α or IL-6 as well as due to secretion from the cells that were damaged¹⁸.

According to a study of Feld et al. the serum level of ferritin was not a good mortality predicting factor during hospital admission having AUC 0.677.15. 799 ng/ml was taken as optimal cut off value. Whatsoever, the increased ferritin level in African American patients were found to be more prone to expire in New York, compared with the results associated with this study. The serum ferritin level was also used to determine the severity of disease and the complications¹⁹⁻²⁰.

The study of Herold et al. reported that AUC 0.750 is needed to predict the ventilatory support with cut off value about 1285 ng/ml. The seven-fold increased CRP is the best to estimate mortality in patients. Various cut off values were presented for CRP to evaluate the death rate and the complication in many studies. According to the metaanalysis of Huang et al, the patients with CRP ≥ 10 mg/L are three times nearer to mortality²¹. According to Liu et al, the CRP > 41.8 mg/L is more associated with severe diseased complications.19 a study of Luo et al. observed CRP > 41.4 mg/L had 90.5% sensitivity and 77.6% specificity in association with mortality in admitted patients²².

The elevated level of LDH is also found in other infections as well. For example, MERS-CoV and H5N1. The ROC curve in this cohort, evaluate two times increased prediction of mortality during hospital stay. An American study revealed LDH levels ≥ 1200 U/L is 8 times nearer to mortality. Hence, the LDH and CRP levels may help clinicians to predict the prognosis²³.

According to the study of Asghar et al., the data from 364 patients having covid infection was analyzed. This study also evaluate the lab findings of three inflammatory markers. Comparing it with the present outcome, all presented a lower AUC. The ferritin level was found to be weakest as predicting factor in this study²⁴. So, it may be due to any co-incidence but in our study serum ferritin was the first priority to predict mortality. Most importantly, in their study, the blood samples to check inflammatory markers was done in first half hour of hospital admission according to the policy presented by the study center. Patients with incomplete data at the time of admission were not included in the study so biasness was prevented. Others studied influence of inflammatory markers during hospital admission, so it was interesting to look at the effects of changes on outcomes during hospitalization²⁵.

Due to financial restriction, authors were unable to monitor inflammatory marker levels like IL-6 or procalcitonin in many patients. Also, the standard method of medical notes documentation was not followed due to high load of work. Therefore, the adverse effects on inflammatory markers like CRP due to different factors were not evaluated. The latter being lower in cirrhosis patients or those taking medication like statins. It can be possible that many patients due to mild disease form, missed for testing during hospital admission. The mortality rate was high in this group as compared to the cohort managed in hospital. However, the results highlight the three inflammatory markers as predicting factors of mortality in hospital. According to the authors, to get refined results, more patients are needed for the study.

CONCLUSION

The serum level of ferritin, LDH as well as CRP are the predicting factors of mortality in patients with Covid-19 infections. The ferritin level was found to be better in this regard. The data was collected at the time of hospital admission, when the clinical spectrum of patients was not much deteriorated. This was an opportunity for the clinicians to take a close look on patients with elevated inflammatory markers and begin proper management or intervention to prevent from bad outcomes.

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