ORIGINAL RESEARCH

A Cross-sectional Assessment of Initial Liver Function Tests between Severe and Non-severe COVID-19 Patients and Its Correlation with In-hospital Outcomes: A Rural-based Single-center Study

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Abstract

Background: Coronavirus disease-2019 (COVID-19) is both an infective and inflammatory disorder. The liver is the principal site for the orchestration of inflammatory cascade after entry of SARS-CoV2 in humans. The study of liver-related parameters has immense potential to understand the pathophysiologic alteration and identify the therapeutic targets related to COVID-19 infection.

Method: A single-center, prospective, cross-sectional, and non-interventional study was conducted on the patients who were COVID-19 positive through RT-PCR test. Comparing and correlating the parameters of liver function test (LFT) between the disease severity group (severe vs non-severe) and also the in-hospital-outcome group (discharge and death) and the assessment of the strength of association (by calculating odds ratio [OR]) of individual LFT parameters for disease severity and in-hospital outcomes.

Results: Among the total number of patients 507 suitable patients were identified. Liver function tests at presentation were considered for the study. Individual parameters in LFT were compared and the strength of association was measured with disease severity and in-hospital outcomes. Disease severity was measured by CURB 65, q SOFA, and AIIMS/ICMR 2021 Clinical severity scores. In-hospital outcomes were defined as discharge or death.

We observed significant differences with respect to serum albumin level, SGOT, SGPT, and albumin globulin ratio between severe and non-severe groups of COVID-19-positive patients. Hypoalbuminemia followed by transaminitis were the commonest abnormalities.

Conclusion: Among the individual LFT parameters, hypoalbuminemia followed by transaminitis were the most frequent abnormalities. The altered liver functions were strongly associated with disease severity and outcomes. Hypoalbuminemia and high transaminase (SGOT and SGPT) levels had strong association with disease severity and outcomes.

Keywords: Coronavirus disease-2019, C-reactive Protein-albumin ratio, Liver function test, Serum albumin, Severity.

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Introduction

SARS-COV2 is a multisystem inflammatory disorder. Among the list of various organ involvements, coronavirus disease-2019 (COVID-19) hepatopathy is not discussed adequately in the literature. Apart from normally analyzed parameters in the liver function test (LFT), the study of various laboratory parameters and chemical mediators during acute inflammation modifying the disease course significantly can give some additional information about the nature and pathogenesis of the disease. C-reactive Protein (CRP) is one such well-known parameter. The coagulation cascade plays a significant role in COVID-19 infection. Anticoagulation along with steroids remains the most important therapy in critical COVID-19 patients irrespective of the waves of the pandemic. The liver plays a significant role in COVID-19 pathogenesis but unfortunately, the area is less discussed even after prolonged suffering from the four waves of the pandemic. This study aimed to compare the various LFT parameters at presentation between two groups of patients namely severe and non-severe based on CURB-65 criteria.[1] and quick Sequential Organ Failure Assessment (q SOFA)[2] and clinical severity score according to the AIIMS/ICMR Joint Task Force guideline (published on 22.04.2021). The parameters considered to calculate the CURB 65 scoring are mental status, blood urea nitrogen, respiratory rate, blood pressure,

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and age. The q SOFA scoring is easier to calculate based on sensorium, respiratory rate, and systolic blood pressure. In one of our previous studies, we observed that CURB-65 had better predictability for disease severity and outcomes than q SOFA.[3] Severity was also graded into three categories—mild, moderate, and severe based on ICMR clinical severity grading. It is based on vital parameters like respiratory rate, peripheral oxygen saturation, and SBP. Hypoalbuminemia is an independent predictor of severity and mortality in COVID-19.[4] An autopsy-based review article by Zanon et al.[5] concluded that postmortem investigation remainder the gold standard to investigate the effects of SARS-CoV2 in different organs. Despite the limited number of autopsies performed worldwide today there is no doubt that the liver is the target organ for the fire despite minimal viral receptor expression how you are not damaged is not always directly linked to the action of the virus but can be secondary to inflammation or even simply caused by therapy administration during hospitalization. Therefore, it is important to monitor the patients, by observing the changes in liver function which can affect the patient's outcome. The liver is the principal site for the commencement of the inflammatory cascade. So, study related to sequential alteration of liver function demands extensive studies in the future. A study conducted by Cai et al. in 2020 found 76.3% had abnormal LFT among 417 COVID-19-infected patients.[6] This study also concluded that patients with abnormal LFT were at higher risk of progressing to severe disease. An editorial letter to the *Journal Liver International* in 2020 mentioned about 21.3% elevation of ALT and a 22.2% elevation of AST. Severe patients showed a higher proportion of AST and ALT elevations[7] and AST was associated with the highest mortality according to the study conducted by Lei et al.[8] An original article by Efe et al.[9] concluded that COVID-19 vaccination may be associated with immune-mediated liver injury, and may be beneficial to corticosteroid therapy.

So, it is obvious from the literature review that there is a wide range of variations in various study results related to this topic. Ongoing studies on the previously investigated areas are always beneficial for a disease like COVID-19 as the virus is a highly mutating one and little is known about its pathogenesis and targeted therapy. This study aims to search the pattern of LFT in a rural-based tertiary care COVID-19 hospital and its association and correlation with disease severity and in-hospital outcomes.

Aims and Objectives

Comparing and correlating the parameters of LFT between disease severity groups (severe vs non-severe) and also in-hospital-outcome group (discharge and death).

Assessing the strength of association [by calculating odds ratio (OR)] of individual LFT parameters with the disease severity and in-hospital outcomes.

Materials and Methodology

Approval from the Institutional Ethics and Scientific Review Committee of DHGMCH was obtained for the study [Memo-DHGMCH/2023/432 dt.14/03/2023]. The study was conducted with the data collected from one-year retrospective data in a Government Medical college in India. This is a descriptive cross-sectional study on retrospective data of RT-PCR-confirmed COVID-19 patients of more than 12 years of age. All the 1,370 admitted RT-PCR COVID-19-positive patients were considered for the study as a target population. Among them, 568 had the required uploaded electronic medical records (EMR) and bed head tickets (BHTs) data. Among them, 507 were selected for analysis after excluding the patients under 12 years of age and obstetric patients as they were expected to have altered body physiology. All the data was collected from the Institutional Medical Record Section after obtaining proper administrative approval.

Table 1: Descriptive statistics of patients

	n	*Median*	*IQR*	*Minimum*	*Maximum*
Age Year	507	52	23	3	100
Length of stay	507	8	8	0	184
EWS	503	7	5	2	26
q SOFA	500	1	1	0	3
CURB 65	461	0	1	0	4
CRP value	230	35	58.4	0	412
BI	507	0.5	0.4	0.14	24.7
Albumin	503	3.33	0.755	0.2	4.9
Globulin	502	3.4	0.7	1.56	5.77
AGR	502	0.993	0.348	0.0588	2.33
SGOT	506	37.5	37.75	12	2564
SGPT	504	37.5	51.75	6	1567
SGOT: SGPT ratio	504	1.023	0.678	0.0902	12.07
ALP	504	81	45	6.7	985
CAR	503	0	8.487	0	158.46

Age, sex, q SOFA score, ICMR disease severity grades, having comorbidities or not, vaccinated or not, serum total bilirubin, serum albumin level, serum globulin level, albumin to globulin ratio (A:G ratio) SGPT (ALT), SGOT (AST), SGOT:SGPT ratio, CRP to Albumin Ratio (CAR) were the various study variables considered for the study. Disease severity was calculated based on three well-established simple scoring systems-CURB-65, q SOFA, and ICMR clinical disease severity grades separately. In-hospital outcomes were defined as discharge or death.

All the collected data were put in a Microsoft Excel sheet and analyzed by Jamovi version 2.3(2022) statistical software. As the dataset showed a skewed pattern, appropriate non-parametric tests were carried out. The numerical data were presented as median and inter quartile range (IQR). The categorical variables are presented by numbers, percentages, and proportions, and associations between the variables were analyzed by Chi-square test. Mann–Whitney *U* test was used to compare between the two groups and the Kruskal–Wallis test was used to compare the numerical values between more than two groups. The strength of association of individual LFT parameters with disease severity and outcomes was analyzed by calculating OR from a 2×2 table. it was considered OR > 1 as a significant strength of association and a p-value < 0.05 has been considered statistically significant at a 95% confidence interval (CI). The LFT values for the patients were taken at the time of the admission and while selecting the patients an appropriate matching was made with respect to the duration of illness.

Results and Analysis

Descriptive Statistics (Table 1)

The median age (year) (IQR) was 52(23). The Median average length of stay (days) (IQR) was 8(0.8). A statistically significant difference was observed in relation to the length of hospital stay between two groups of patients with or without altered LFT (p-value < 0.01). The median

Table 2: Mann–Whitney *U*–q SOFA severity score

	Statistic	*p*
BI	7495	0.792
Albumin	4575	<0.001
Globulin	7094	0.688
AGR	5793	0.039
SGOT	5990	0.034
SGPT	7385	0.734
SGOT:SGPT Ratio	4890	<0.001
ALP	7408	0.756
CAR	7158	0.711

Table 3: Mann–Whitney *U*-outcomes

	Statistic	*p*
BI	13852	0.262
Albumin	8646	<0.001
Globulin	13915	0.552
AGR	9995	<0.001
SGOT	11075	<0.001
SGPT	12750	0.063
SGOT:SGPT Ratio	13085	0.12
ALP	12734	0.043
CAR	12376	0.028

length of hospital stay was 9 days in the altered LFT group where whereas it was 7 days for patients without alteration of LFT.

Among the total 507 admitted patients, 57% were male and 43% were female; 61.1% of males and 60.5% of females had altered LFT.

The median q SOFA score (IQR) was 1.0 (1.0). The median serum bilirubin level (mg/dL) (IQR) was 0.5 (0.4). The median serum albumin and globulin levels (mg/dL) (IQR) were 3.3 (0.75) and 3.4 (0.7), respectively and the AGR (IQR) was 0.993 (0.348). The serum SGOT (IU/L) (Median, IQR), SGPT (IU/L) (Median, IQR), and SGOT:SGPT ratio [Median, (IQR)] were 37.5 (37.5), 37.5 (51.75), and 1.023 (0.678), respectively. The median Alkaline phosphatase level (IU/L) (IQR) was 81 (45.0). The median (IQR) CRP value (mg/dL) and CAR were 35.0 (58.4) and 0.0 (8.4), respectively.

Alteration of LFT was observed in 61% of patients when we considered collectively. Note that 39% of patients had normal LFTs. The majority (56.2%) of the LFT alteration was observed in the 31–60 years of age group (*p*-value 0.012). Also, 60.3, 60.9, and 60.7% of patients had altered LFT when disease severity was calculated according to CURB 65, q SOFA, and Clinical severity grading, respectively. 83.7% of clinically severe disease had altered LFT (*p*-value < 0.001) (Table 2). A total of 81.8 and 72% had altered LFT when severity was calculated on the basis of q SOFA (*p*-value = 0.011) and CURB 65, respectively (*p*-value is 0.221).

Also, 60% of the patients had hypoalbuminemia (serum albumin <3.5 mg/dL) and 37% of the patients had high globulin levels (>4.5 mg/dL). We found that 20% of patients had altered AGR (<1), 35% of the patients had mildly raised SGPT (within three times of upper limit of normal value), and 11% had significantly raised SGPT (more than three times of upper limit of normal value). So, 36.8% of patients had raised SGPT level, 43.8% of patients had significantly raised (more than three times of upper limit of normal value) SGOT level, and 8.6% had mildly raised SGOT (within three times of upper limit of normal value). It was also found that 45.4% of the patients had high SGOT levels, 49% had ALT to AST ratio less than one (<1) and 42.9% had a ratio of 1–2. Only 8.1% had an ALT to AST ratio of more than 2 (>2) and 87.3% had normal ALP value. 11.3% had mildly raised alkaline phosphatase (ALP) (within three times of upper limit of normal value) and only 1.4% had significantly raised ALP (more than three times of upper limit of normal value). So, only 12.7% had raised ALP value.

In total, 86% of patients where survived and 14% of patients were succumbed. 60.8% of patients had altered LFT who succumbed (*p*-value < 0.001).

Table 4A: Contingency tables—altered LFT and clinical severity

	LFT altered or not		
Clinical severity grade	*Altered*	*Normal*	*Total*
Mild			
Observed	189	158	347
% within row	54.5%	45.5%	100.0%
Moderate			
Observed	78	31	109
% within row	71.6%	28.4%	100.0%
Severe			
Observed	36	7	43
% within row	83.7%	16.3%	100.0%
Total			
Observed	303	196	499
% within row	60.7%	39.3%	100.0%

Table 4B: χ^2 tests

	Value	*df*	*p*
χ^2	20.6	2	<0.001
N	499		

Analytical Statistics

When we compared the medians of various non-parametric variables between severe (q SOFA > 2) and non-severe (q SOFA < 2) groups by Mann–Whitney *U* test (Table 3), we found a statistically significant difference with respect to Albumin level (*p*-value < 0.01), AGR (*p*-value < 0.01), SGOT/AST level (*p*-value < 0.01) and SGOT/SGPT ratio (*p*-value 0.003). Similar results were observed when we compared the disease severity by the Kruskal–Wallis test (Tables 4A and B) based on Clinical severity as per ICMR guidelines which is based on peripheral oxygen saturation (SpO2), systolic blood pressure, and respiratory rate. ICMR Disease severity had three levels–mild, moderate, and severe.

Again, when we compared the medians of various non-parametric variables between discharge and death that is, outcome groups (Table 5), similarly we found a statistically significant difference with respect to albumin level (*p*-value < 0.01), AGR (*p*-value < 0.01), SGOT/AST level (p-value < 0.01). Along with the

Table 5: Kruskal–Wallis results

	χ^2	*df*	*p*
BI	2.25	2	0.325
Albumin	37.49	2	<0.001
Globulin	1.24	2	0.537
AGR	20.42	2	<0.001
SGOT	8.83	2	0.012
SGPT	5.44	2	0.066
SGOT: SGPT Ratio	11.34	2	0.003
ALP	3.54	2	0.171
CRP value	3.16	2	0.206
CAR	5.47	2	0.065

Table 6A: Contingency tables—albumin level and CURB-65 severity

	Albumin level		
CURB65 severity	*Hypalbuminaemia*	*Normalalbuminemia*	*Total*
ICU Admission			
Observed	19	4	23
% within row	82.6%	17.4%	100.0%
Ward admission			
Observed	256	178	434
% within row	59.0%	41.0%	100.0%
Total			
Observed	275	182	457
% within row	60.2%	39.8%	100.0%

median differences we also observed a statistically significant difference with respect to ALP level (*p*-value 0.043) and CAR (*p*-value 0.023). When we compared the median values of various LFT parameters between the discharged and death group, we found again a statistically significant difference with respect to albumin, SGOT, and SGPT where the *p* values were less than 0.01 at 95% CI.

On analysis of the categorical variables by Chi-square test, we couldn't observe any significant difference in LFT alteration in patients with added risk factors or prior history of COVID-19 vaccination. A strong association was observed [OR 3.10 (1.65–5.84)] between altered LFT and mortality (*p*-value < 0.001). When the association between altered liver function and disease severity by CURB-65 criteria, we observed a significant OR [3.30 (1.10–9.87)] but we couldn't observe a significant association when disease severity was calculated by q SOFA score (OR 0.194) though the statistically significant differences were present in both the scenarios (*p*-values were 0.024 and <0.001, respectively) at 95% CI.

Hypoalbuminemia had significant association with both disease severity by CURB-65 [OR 3.30 (1.10–9.87)] and outcomes [OR 5.17(2.50–10.7)] (Tables 6A and B). In addition, a strong association with hypoalbuminemia was absent when we considered the q SOFA-based disease severity scoring. The same influential strength of association was observed with altered AGR as serum albumin level is a significant component of the ratio. As ICMR clinical disease severity had three categorical grades, OR couldn't be calculated though a statistically significant difference was obtained when compared against altered liver function status (*p*-value 0.008 at

Table 6B: χ^2 Tests

	Value	*df*	*p*
χ^2	5.09	1	0.024
N	457		
Comparative measures			
		95% Confidence intervals	
	Value	Lower	Upper
Odds ratio	3.3	1.1	9.87

95% CI). Statistically significant differences were observed when we compared the disease severity and outcomes with serum SGPT (*p*-value < 0.01) and SGOT values (*p*-value 0.023).

Discussion

The study was conducted in a dedicated COVID Hospital attached to a rural-based Medical College in the Southern part of West Bengal in the Sundarbans area. In brief, Initially, it was an 80-bed COVID Hospital with 15 critical care beds. Later on, during the second wave of the pandemic it was converted to 250 bedded COVID Hospital during the second pandemic wave. A study conducted by Grando et al.[10] concluded that liver damage due to COVID-19 is very common, especially with critical disease. This finding is also more pronounced in patients with pre-existing chronic liver disease. Direct infection, immune-mediated injury, DILI, and hypoxic injury were proposed mechanisms of action for liver injury in COVID-19 patients. They also reminded the necessity of a large cohort study to determine the long-term effect of COVID-19-induced liver injury.

A retrospective analysis of 105 patients conducted by Wang et al.[11] observed that elevated liver function index is very common in patients with COVID-19 infection but moderate elevations were common and mostly reversible.

According to this study, the Median age of patients in this study was 52 (23) yrs. Among them 57% were male and 43% were female. This proportion of sex distribution was similar to the study.[12] It is observed in this study that 61% of patients had some form of abnormal LFT at admission. A single center-based study conducted by Wang et al.[11] from Beijing observed that 56.2% had abnormal LFT. In a research article published in the *Journal of Hepatology* by Cai et al. [6] on 417 patients, 76.3% had a normal liver test and 21.5% and liver injury during hospitalization. The patient with abnormal liver test had a significantly higher chance of developing severe COVID-19 pneumonia. We observed that abnormal was almost a 60% association with disease severity. A study conducted by Hundt et al.[12] in the *Journal Hepatology* also concluded abnormal LFT was found in most of the hospitalized patients and associated with poor clinical outcomes. In this study, we observed almost 61% of patients had abnormal LFT. Similar observations were noticed in this study. We observed significant differences between survived and succumbed groups with respect to hypoalbuminemia, high SGOT, and SGPT levels. According to our study results, 86% of patients where survived, 14% of patients succumbed, and 60.8% of patients had altered LFT who succumbed *(p*-value < 0.001).

A mini-review published in the *World Journal on Gastroenterology* by Hu W et al.[13] in December 2022 concluded that the risk factors for liver dysfunction were male sex, older age, and comorbidities like diabetes, hypertension, and obesity. Any pre-existing liver disease

was identified as a potentially significant risk factor for liver injury in COVID-19 patients. Collectively, liver injury in COVID-19 was associated with severe infection and required additional attention and effective treatment. They also recommended targeted therapy for aged patients with liver injury and attention should be paid to identify the existence of DILI. Surprisingly, we couldn't observe any significant difference of LFT alteration in patients with added risk factors or prior history of COVID-19 vaccination.

We observed that there is a significant difference in hospital stay duration between patients with and without altered LFT. In a review article by Cichoż-Lach and Michalak[14] It is observed that impaired LFT parameters are common findings in COVID-19 infection and may affect almost one-third of the patients. Deterioration of liver function prolongs the duration of hospitalization. Abnormal LFT can be a predictor of disease severity. Most male elderly with higher BMI had abnormal LFT. Drug-induced liver injury were more common. Patients with CLD had more severe disease course. We considered the initial LFT parameters. Long-term follow-up is required to comment on this and failing to do so is a limitation of our study.

A comprehensive review and meta-analysis conducted on 128 subjects published in *Hepatology International* in 2020 by Kumar Praveen M et al.[15] concluded that the most frequent LFT abnormality was found to be hypoalbuminemia followed by GGT and transaminase abnormalities. Similar findings were observed in our study. In our study, we observed that 60% of the patients had hypoalbuminemia and around 43–44% had transaminitis. Moreover, we observed hypoalbuminemia is an independent risk for disease severity and mortality. This finding is also similar to the findings revealed in a study.[11] This study also claims that hypoalbuminemia is caused by hepatotoxicity due to cytokine storm. An article published in in 2022 *Journal of Clinical Medicine* authored by Tokarczyk et al.[16] on 401 patients observed that albumin is a better predictor of the COVID-19 severity compared to neutrophil lymphocyte ratio (NLR). They also mention that hypoalbuminemia with AST or total bilirubin was found to be an excellent predictor of in-hospital mortality which shows the recommended utility of early evaluation of serum albumin for early severity assessment.

A review article by Amin M[17] concluded that severe COVID-19 infections developed important alterations in liver enzymes. We found along with albumin level serum transaminase levels were altered in around 43–44% of cases. However, no significant alteration of serum ALP level was observed and ALP levels were not associated significantly with respect to disease severity and in-hospital outcomes. A review article in *Digestive and Liver Disease* by Metawea observed almost 50% of patients had abnormal LFT. This article also mentioned about normal alkaline phosphatase (ALP) almost in all patients.[18] In a review article by Tian and Ye,[19] it is observed that COVID-19 combined liver injury is common and it correlated with disease severity. Various mechanisms were mentioned as responsible for liver injury. It also observed a correlation with disease severity. A similar finding was observed in this study.

In this study, we observed, 45.4% of patients had high SGOT levels and 46% of patients had raised SGPT. In a specially reviewed paper with a special focus on pregnancy by Cooper et al.[20] demonstrated AST predominant transaminitis in COVALI associated is with written poor outcomes. Though we had not included obstetric patients in this study, we found similar observations in relation to SGOT levels in non-pregnant patients. Another review article by Li et al.[21] concluded that 19 associated liver injuries are caused by the cumulative effect of multiple factors and most patients have transient elevation of liver enzymes. Long-term follow-up studies are invited to get familiar with etiologias of COVID-19-associated liver injury. In a systemic review and meta-analysis conducted by Bzeizi et al.,[22] it was observed that no major changes in serum bilirubin and transaminase levels. But unfortunately, they considered mostly case reports and observational studies. The observation in our study is discordant with the findings of the above-mentioned study.

Here in this study, we only considered the initial LFT on admission to correlate with disease severity and outcome. A sequential follow-up would have been better to comment on progressive changes in LFT values and the effect of various factors on the liver. This is one of the major limitations of the study.

The collection of robust data from EMR and BHTs by trained data entry staff and considering all the available patients for analysis were the major strengths of the study.

Conclusion

The liver can be affected in many ways during COVID-19 infection and it is one of the principal sites for initiation of inflammatory cascade. It is observed from this study that a substantial percentage of patients had liver function abnormalities. Measuring disease severity by CURB 65 or ICMR clinical severity correlates better than q SOFA scoring. Among the individual LFT parameters, hypoalbuminemia followed by transaminitis were the most frequent abnormalities. The altered liver function was strongly associated with disease severity and outcomes. Hypoalbuminemia and high transaminase (SGOT and SGPT) levels also had a strong association with disease severity and outcomes. Besides common LFT parameters, other parameters related to liver pathology and inflammatory cascade can give potential clues for future therapeutic success. So further longitudinal studies on COVID hepatopathy can illuminate the gray areas related to the understanding of the pathogenic mechanisms of this mysterious infection.

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