Pacific Journal of Medical and Health Sciences
ISSN: 2456-7450 | Vol.4, No.4, 2022, pp - 13-53

PJMHS
PACIFIC JOURNAL OF MEDICAL AND HEALTH SCIENCES

Research Paper

Evaluation of the clinical features, radiological findings and inflammatory markers in Covid-19 patients and their corelation with diseaes progression and outcome - A prospective, cross-sectional observational study

Japan Patel[1]*, K.R. Sharma[2], Jagdish Vishnoi[3]

[1]PG Resident, [2]Professor and Head, [3]Professor

Department of General Medicine, Pacific Medical College and Hospital, Udaipur, Rajasthan, India

***Corresponding author Email:** japanpatel24@gmail.com

ABSTRACT

A prospective, cross-sectional observational study was conducted on 167 RT-PCR positive COVID-19 patients admitted to a tertiary care hospital in Udaipur. They were evaluated clinically, radiologically and biochemically and correlated with disease progression and outcome. Clinically, they were categorized as mild (42), moderate (72) and severe (53) with male dominance (71%). Diabetes and hypertension were common co-morbidities. Out of all inflammatory markers studied, D-dimer and Ferritin had significant correlation at any point of time for prognosis while IL-6 was significant for outcome and prognosis in ICU patients. LDH demonstrated significant early trends for outcome while 48 hourly trends were significant for disease progression. Procalcitonin was significantly associated with disease progression. CRP, however, was inconsistent in all aspects. High HRCT scores were related for longer oxygenation and hospital stay. Clinical parameters and inflammatory markers were more significant then imaging alone for disease progression and outcome.

KEYWORDS: Inflammatory markers, HRCT score, Procalcitonin, Neutrophil-to-lymphocyte ratio (NLR)

INTRODUCTION

At the End of 2019, a new respiratory tract virus was discovered in south-east China in the city of Wuhan named SARS-Cov-2, A novel coronavirus later named COVID-19. The study of Genome Sequencing and Phylogeny interprets as 2019-nCoV is a form of Beta coronaviruses associated with human severe acute respiratory syndrome (SARS) and Middle East respiratory syndrome (MERS)[1,2].

2019-nCov are found to be similar to Bat coronavirus, and it is put forward that bats are the primary sources, But the origin of the 2019-nCoV is still being inquired, current attestation indicates the spread to humans occurred via dissemination from inhospitable animals unlawfully sold in Huanan Seafood Wholesale Market[3].

The Virus spread expeditiously through China infecting more than 1,40,586 people till January 20224. In a short period, it took over Europe causing huge loss of life in Italy, Spain, France, Germany, the UK, and the USA. The WHO declared COVID-19 a Global pandemic on 11th March 2020[5].

COVID – 19 spreads to India in late January 2020, and within the span of 4 weeks, the cases surged to a few

hundred numbers[6]. As of January 2022, there were 37,69,41,709 confirmed cases worldwide and around 56,95,604 confirmed deaths[7]. In India there were around 4,21,88,138 confirmed cases of covid-19 with 5,01,979 confirmed deaths till 31st January 2022[8].

Before 3rd March 2020, India had only 3 cases of covid-19 from the state of Kerala all of them were treated and discharged. On 3rd March 2020, India's 4th case was found at Jaipur in the State of Rajasthan, and found later that this patient had infected 17 other Italians who were on a tour to Rajasthan, India[9]. There were 12,00,052 cases in Rajasthan as of Jan-2022[10].

SARS-COV-2 is transmitted from human to human via means of contact in terms of salivary-respiratory droplets which enter the body through the mouth or nasal cavity[11]. After the entry of SARS-COV-2, the infectious process starts when the organism passes through the mucosal membrane of the larynx and nose, which enters the Upper Respiratory Tract, the first symptom includes fever, myalgia, anosmia, cough, and hypogeusia[12]. Lungs are the primary organs affected by SARS-COV-2 infection and later on other systems namely heart, gut, kidney, liver, brain, spleen, and lymph nodes can be affected. Higher ACE2 expression in these tissues will correlate with disease severity and will potentiate multiorgan failure[13,14]. In the initial stage, viral load is higher in Lower Respiratory Tract but later on in terminal stages, pulmonary edema fills up the alveoli with Hyaline Membrane formation and leads to increased mortality and morbidity[15].

COVID–19 mimics a pneumonia like illness, having an extremely varied clinical picture from asymptomatic patients to symptomatic patients having mild, moderate, or severe courses of the disease[16] The clinical course of the disease is related to numerous risk factors that contribute to mortality[17-21].

Effective biomarkers can play a crucial role in identifying, managing, and preventing severe complications in patients with COVID-19 due to the rapid progression of the disease[22]. Some biomarkers that are commonly evaluated to assess the severity of COVID-19 include D-Dimer, serum ferritin, C-reactive protein (CRP), interleukin-6 (IL-6), and lactate dehydrogenase (LDH).

D-Dimer is a marker of hypercoagulability, and its elevated levels have been linked to the progression of COVID-19[23]. CRP, a marker of inflammation[24] and LDH, which is released from lung tissue in greater amounts in severe COVID-19 infections and can indicate a severe form of interstitial pneumonia[25]. The activation of the immune system in response to the virus leads to the release of a high number of cytokines, one of which is IL-6. IL-6 is a versatile cytokine that has both anti-inflammatory and pro-inflammatory effects and plays a role in the body's defence against infection[26]. However, excessive production of IL-6 while fighting the virus can lead to a serious systemic inflammatory response known as a cytokine storm. Research has shown that measuring the levels of IL-6 in the plasma and/or bronchoalveolar fluid can serve as an early indicator of lung injury and predict the likelihood of prolonged mechanical ventilation, organ dysfunction, and poor outcomes in lung diseases[27]. Elevated levels of ferritin, which is a degradation product of haemoglobin, have been found to be associated with poor outcomes in COVID-19 patients[28].

Radiographic imaging is the most significant step in identifying the severity of the disease. Chest X-Ray can be useful in primary triaging of the disease. CT scanning can help identify lung involvement and the extent of the infection, especially in cases where Clinical and Radiographic pictures of the disease do not match[29].

Inspite of the debatable argument, that a CT scan cannot be used as a routine tool for screening purposes[30-39] there is no denying that a CT scan is extremely reliable in evaluating the extent and spread of the COVID–19 disease in lung and help in giving a numerical grading system for better categorizing the disease severity[35,36].

A study by Li *et al.* described that the clinical picture of the COVID–19 disease is a direct extent of the visual (quantitative) evaluation of Radiographic (specifically CT) findings. These findings include Bilateral pulmonary parenchymal Ground-Glass and consolidative opacities[33,40] CT Scan, thus quantify the underlying or residual lung parenchyma which can also help in understanding and estimating alveolar recruitment, during the ventilation process. It can also help patients suffering from acute respiratory distress syndrome, by evaluating the prognosis[37].

SARS COV-2 consists of 4 structural proteins, namely surface glycoprotein (S), small envelope protein (E), matrix protein (M) and nucleocapsid protein (N). The S gene codes for the receptor-binding spike protein which ultimately ends up in the infection of the Virus[41]. There have been RT-PCR kits developed to detect genes of SARS-COV -2. This procedure involved the reverse transcription of SARS -CoV – 2 RNA into the cDNA strands, ending with the amplification of specific cDNA regions. The E gene (envelope protein gene) and the N (nucleocapsid protein gene) and RdRP gene (RNA dependent RNA polymerase gene) are the regions having conserved sequences. RT-PCR hence is the most predominantly used method for diagnosing COVID-19 using respiratory samples[42].

For the cyclic threshold value, Ira Praharaj *et al.* studied a total of 110 samples each of 5 and 10 sample pools. They concluded that concordance between 5 sample pool and individual sample testing was 100 percent in C_t values $\leqslant$30 cycles and 95.5 percent for C_t values $\leqslant$ 33 cycles[43] the present study was undertaken to evaluate the clinical characteristics, biomarker levels, and the severity of COVID-19. Additionally, the study also aimed to correlate these findings with high-resolution computed tomography (HRCT) chest scans to identify patients at risk of fatal complications.

MATERIALS & METHODS

The present study was conducted on RT-PCR confirmed COVID-19 patients who were admitted to department of

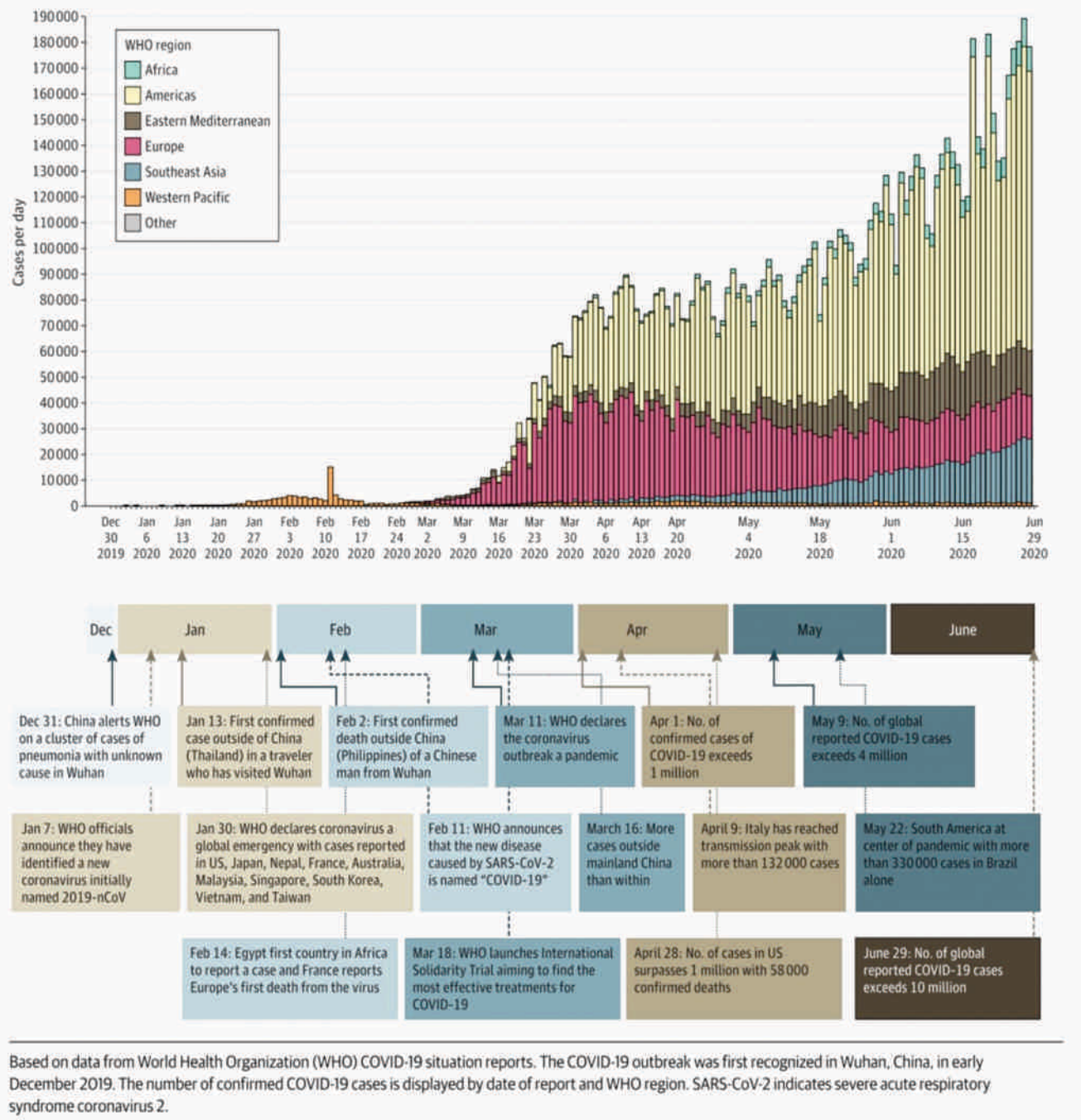


Figure A: Key Events in the Early Coronavirus Disease 2019 (COVID-19) Pandemic[44]

general medicine in Pacific Medical College and Hospital, Udaipur, Rajasthan from December 2020 to May 2021. Patients with only rapid antigen test positive for COVID-19, probable cases in which findings were suggestive of covid-19 on HRCT scan of the chest but with negative PCRs for SARS-COV-2 on 2 consecutive occasions, pediatric patients and pregnant females were excluded from the study. It was a prospective, cross-sectional observational study approved by Institutional Ethical Committee vide reference number PMU/PMCH/IEC/PG/2020/25. Patients were enrolled after written informed concern and blood samples were collected within the first half hour of admission for tests including: quantitative CRP, serum ferritin, and serum LDH. Data on these laboratory parameters as well as the duration of hospital and ICU stay, Mode and duration of oxygenation, and Important Treatment given data including antiviral, heparin, steroids, Convalescent plasma, and Immunomodulator drugs, the primary outcome were collected. Disease severity was assessed using Clinical Guidelines from the Ministry of Health & Family Welfare, Government of India (MoHFW, Gov. INDIA) as given in table-1.

Table 1: Clinical Severity of Covid-19 Infection (adapted from MoHFW, Gov. INDIA)

Category	Definition
Mild	Upper respiratory tract symptoms and/or fever without shortness of breath or hypoxia
Moderate	Any one of the following: 1. Respiratory rate $\geqslant$ 24/min, breathlessness 2. SpO_2 : 90% to $\leqslant$ 93% on room air
Severe	Any one of the following: 1. Respiratory rate >30/min, breathlessness 2. SpO_2 < 90% on room air

According to the disease severity and clinical outcome, these patients were also divided into three groups: Nonsurvived, Survived severe, and Nonsevere for evaluation.

All the patients were treated and monitored following the guidelines issued by the Ministry of Health & Family Welfare, Government of India.

General treatments included supportive therapy, anti-viral agents (eg, Remdesivir, Favipiravir, Oseltamivir) and oxygen supplementation with respiratory support, anticoagulants, and anti-inflammatory or immunomodulatory therapy. Clinically Critically ill or moderate to severe patients were admitted to an intensive care unit (ICU) and supported by intubation and mechanical ventilation if required. In accordance with the criteria stated in the clinical guidelines for hospital management and discharge of a COVID-19 patient.

Data Collection

Patients' demographic and baseline characteristics, symptomatic and radiological characteristics as well as the laboratory findings on admission and follow-up data of laboratory parameters in all Non-severe, Severe & Survived and Non survived patients were obtained and analyzed.

Parameters

1. Clinical Profile: history, symptoms, age, gender, vitals, past medical illness and comorbidities
2. Radiological Parameter: HRCT for the evaluation of lung involvement and radiological findings The available chest CT images of each patient were reviewed by a senior radiologist blinded to the clinical data, in order to confirm detailed abnormality of radiological characteristics of these patients. The presence or absence of the these following features was recorded for each patient: GGO, IST, CP, FB, PF. In Some Moderately ill patients with a prolonged duration of O_2 dependency (>21 days), Repeat HRCT was also done, and those findings were also taken.
3. Laboratory Parameters: All Routine Blood investigations including CBC, RBS, HbA1C, Serum Electrolyte, and Inflammatory markers like IL-6, D-Dimer, CRP (Quantitative), Serum Ferritin, LDH, Procalcitonin, ESR Some of the laboratory parameters and inflammatory markers were also repeated within 48 hours.
4. The clinical outcome of each patient (i.e., nonsurvived, discharged, or remained in hospital) till final follow-up were obtained.

STATISTICAL METHODS:

Data were analyzed using The jamovi project (2022). *jamovi* (Version 2.3) [Computer Software] for windows. Retrieved from https://www.jamovi.org[45]. Qualitative data were expressed as numbers and percentages. Quantitative data were described as mean ± standard deviation. Median and interquartile ranges were used for continuous variables with a non-parametric distribution.

The normality of data was checked using the Shapiro-Wilk test. Kruskal-Walli's test was done to compare median and interquartile ranges for different inflammatory markers amongst patients with mild, moderate, and severe disease.

Amongst the two groups based on outcomes, levels of inflammatory markers were compared using Independent-samples Mann-Whitney U-test. Proportions of patients with elevated levels of inflammatory markers amongst different groups were compared using Chi-square/ Fisher's exact test. $p<0.05$ was considered significant.

Logistic Regression Analysis

All variables were subject to univariate logistic regression, and odds ratios (ORs) were calculated between nonsurvived and survived severe groups, with a 95% confidence interval. Variables were included in binary logistic regression if corresponding P value was less than .05. Binary logistic regression analysis was used to develop a multivariate model to determine the risk factors of death among critically ill patients.

The Receiver Operating Characteristic (ROC) curve was done wherever applicable. For all comparisons, Pearson correlation coefficients were calculated for increasing inflammatory marker levels and lung involvement in HRCT chest to assess the strength of association.

HRCT CHEST

HRCT chest has been performed using 5 mm slice thickness cuts in axial planes.

IMAGING FINDINGS:-

Diffuse ground glass haziness and consolidation with interstitial septal thickening (crazy paving) and bronchiolar & vascular dilatation are seen involving bilateral lungs predominantly along peripheral part of bilateral lower lobes.

CT Severity	% Involvement	Score
Right Upper Lobe	50-75 %	4
Right Middle Lobe	75-100 %	5
Right lower Lobe	75-100 %	5
Left Upper Lobe	75-100 %	5
Left lower Lobe	75-100 %	5
TOTAL		**24**

Score	CT Severity
≤8	Mild
9-15	Moderate
>15	Severe

Few subcentimeter sized lymph nodes are seen involving pre paratracheal, carinal and subcarinal region.

Rest of lungs show normal architecture attenuation.

Trachea is central. Tracheal bifurcation is defined.

No pleural effusion / thickening present.

Degenerative changes are seen in the form of marginal osteophytes in visualized skeleton.

Note is made of Cholelithiasis.

IMPRESSION:

- ***Suggestive of atypical viral pneumonia (CO-RADS – 5) with CT severity score 24/25. Advise: Further evaluation with RT-PCR.***

Sample HRCT Plate and HRCT Report

Table 2: Demographics and Clinical Features of COVID-19 Patients Classified as "Mild"," Moderate", and "Severe"

Clinical Variables		Mild (n =42)	Moderate (n =72)	Severe (n=53)	p Value
AGE, mean (SD), y		50.1 (14)	52.9 (14.2)	56.9 (13.7)	< .001
Gender	**Females (%)**	14	22	12	0.47
	Males (%)	28	50	41	
Co-morbidities					
Hypertension		10	21	16	0.765
Diabetes (Known case)		10	17	14	0.929
Diabetes (Newly diagnosed)		3	8	11	0.118
Renal dysfunction		0	9	0	
CAD		1	5	0	
CVA		2	1	2	
Hyperthyroid		0	2	0	
Hypothyroid		3	7	0	
Malignancy		2	0	1	
others		3	4	9	
Vitals on Admission					
Pulse Rate (bpm)		85.29 (12.21)	90.32 (16.71)	94.42 (18.99)	0.276
Respiratory Rate(/min)		19.71 (2.12)	22.71 (2.88)	25.08 (6.84)	< .001
Systolic Blood Pressure (mm/hg)		128.83 (19.13)	124.81 (15.2)	132.15 (17.87)	0.078
Diastolic Blood Pressure(mm/hg)		80.48 (10.41)	75.89 (10.82)	78.64 (12.42)	0.975
SpO2(%)		95.71 (1.49)	93.81 (2.46)	83.58 (8.26)	< .001
Random Blood Sugar (mg/dl)		177.97 (115.67)	208.45 (120.77)	223.36 (113.37)	0.298

A total of 167 patients with covid-19 were evaluated using different demographic, laboratory, and radiological parameters. There were 42 patients classified as "Mild", 72 as "Moderate", and 53 as "Severe". The mean age of patients in the "Mild" category was 50.1 years, while the mean age of patients in the "Moderate" and "Severe" categories was 52.9 years and 56.9 years, respectively($P < 0.001$) there were no significant difference in gender distribution among the three categories, males being the majority in all three categories. (figure-1 and table-2).

In terms of co-morbidities, Diabetes was the most common in all categories, followed by hypertension. Renal dysfunction was more common in the "Moderate" category. Only a small number of patients had CAD, CVA, hyperthyroidism, hypothyroidism, or malignancy.(Table-2).

In terms of vitals on admission, the "Mild" category had a lower respiratory rate and higher SpO_2 compared to the "Moderate" and "Severe" categories, and the difference was statistically significant ($p<0.001$). The "Severe" category had a higher respiratory rate and a lower SpO_2 compared to the other categories. The systolic blood pressure was higher in the "Severe" category compared to the "Moderate" category, but the difference was not statistically significant. In addition, the duration of oxygen requirement was significantly longer for the severe group. The other vital signs did not differ significantly among the categories.(Table-2)

Regarding the treatment, the most common treatments given to all groups were Remdesivir, Ivermectin, and Heparin. Steroids were given to a higher percentage of moderate and severe patients compared to mild patients. Plasma therapy was also given more frequently to the moderate and severe groups. The median length of hospital stay was significantly longer for the severe group. As expected, the mortality rate was higher in the severe group, with 13 deaths, compared to 6 in the moderate group and 1 in the mild group. (table-3)

On analysis of symptoms in 167 COVID-19 patients, it was evident that the most common symptoms reported were cough (77.8%), fever (76.6%) and shortness of breath (57.5%). Easy fatigability was reported in 23.4% of the patients, while only 8.4% reported body aches. Only less than 10% of the patients reported other symptoms, which were decreased Appetite, sore throat, vomiting, chest pain, diarrhoea, anosmia, and loss of taste.(fig-2)

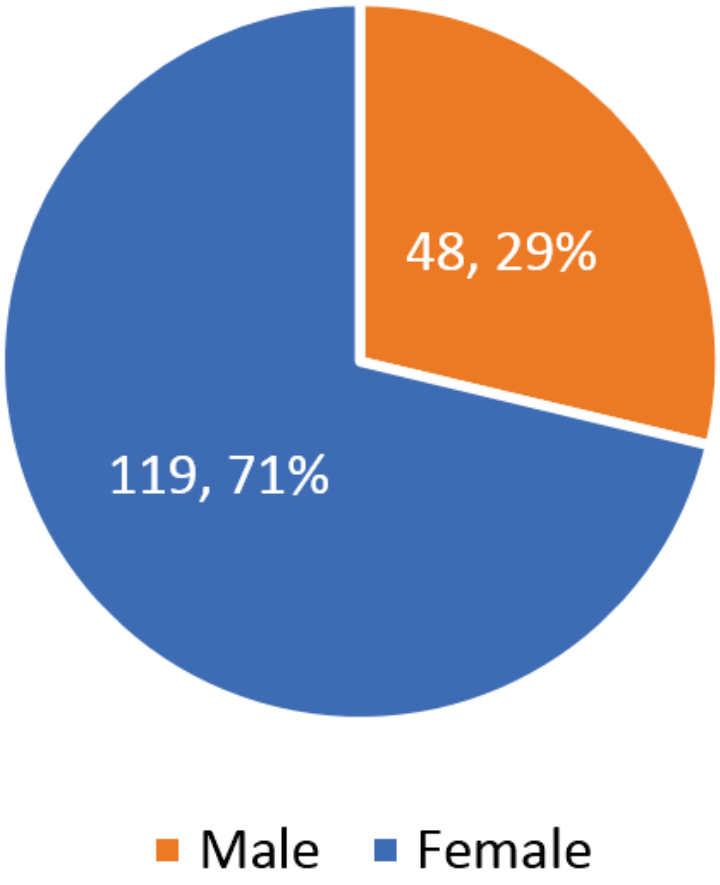


Figure 1: Gender distribution

Table 3: Treatment, Mode and requirement of Oxygenation, Treatment given in COVID-19 Patients Classified as "Mild"," Moderate", and "Severe"

Clinical Variables	Mild (n =42)	Moderate (n =72)	Severe (n=53)	p Value
Oxygen Requirement on Admission				
HFNC	0	0	1	0.339
HM	0	31	11	< .001
NIV	0	0	2	0.113
NRBM	0	0	2	0.113
R/A	42	41	37	< .001
Total Duration of O_2 during the hospital stay in days				
	4.67 (7.89)	8.31 (8.06)	14.08 (12.13)	< .001
Treatment				
Remdesivir	18	58	50	< .001
Ivermectin	36	53	26	< .001
Heparin	28	65	49	< .001
Plasma	1	9	15	0.002
Tocilizumab	0	0	1	0.339
Steroids	33	70	49	0.003
Length of Stay (in Days) Median (IQR)				
	8 (6 -12)	9(8 -13)	14(10 -20)	<0.001
Outcome				
Discharged	41	66	40	**0.002**
In Hospital Mortality	1	6	13	

HFNC-High flow nasal cannula, HM- Hudson's mask, NIV- Non-invasive ventilation, NRBM- Non-rebreathing Mask, R/A- Room Air

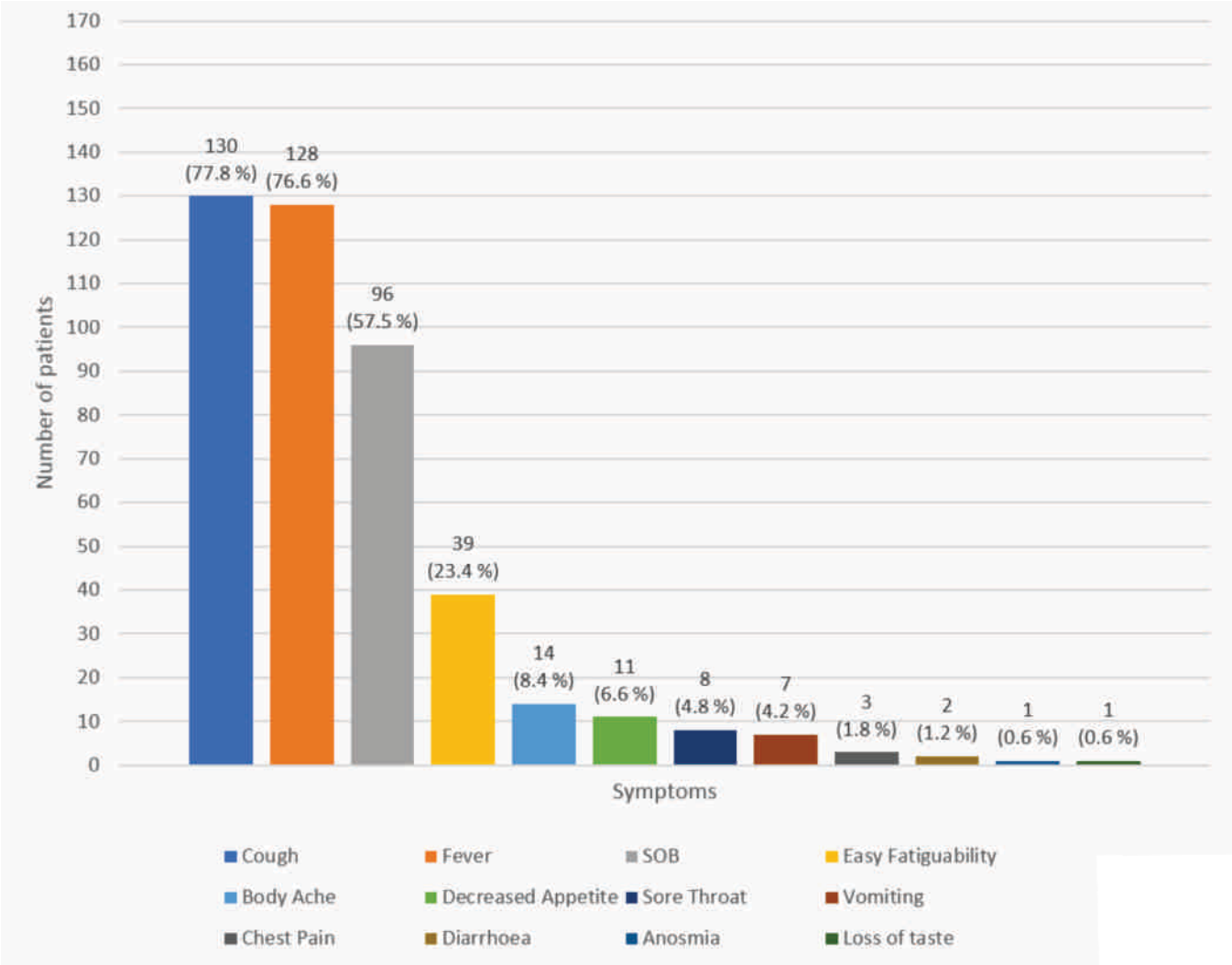


Figure 2: Distribution of Symptoms

Results of pearson's correlation indicated that-

1. There was a significant (r(165)= 0.386, p< 0.001) medium positive relationship days on oxygene and HRCT score on admission (fig-3)
2. A significant r(165)=0.288, p<0.001) small positive relationship between total hospital stay and HRCT score on admission(fig-4)
3. A significant (r(165)= 0.349, p<0.001) medium positive relationship between days in ICU and HRCT score on Admission(fig-5)
4. A non-significant (r(165)=0.136, p=0.080) very small negative correlation between total ward stay and HRCT score on admission(fig-6)

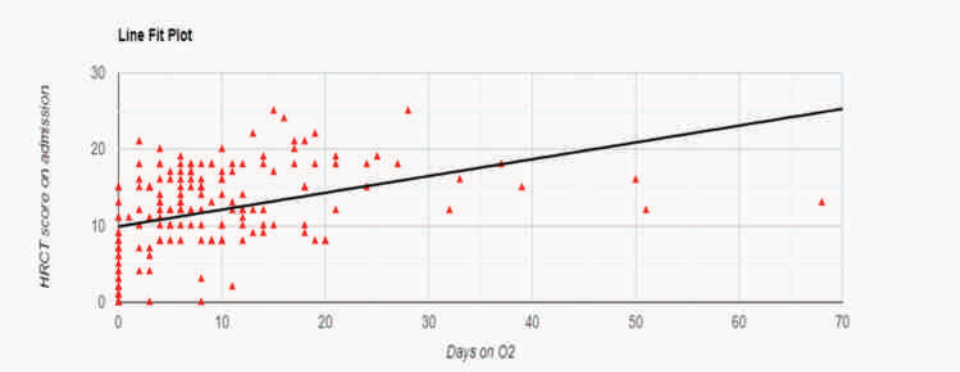
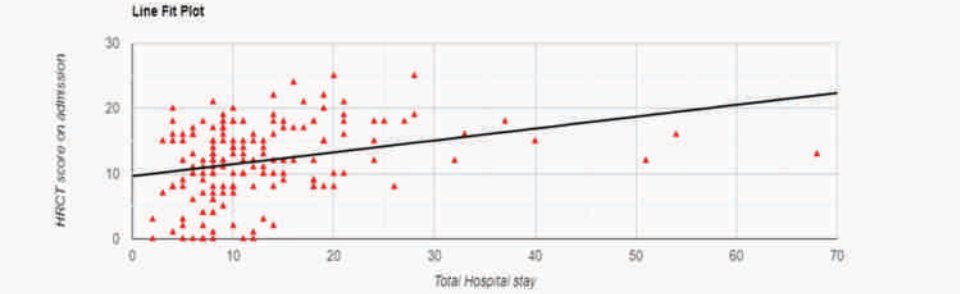
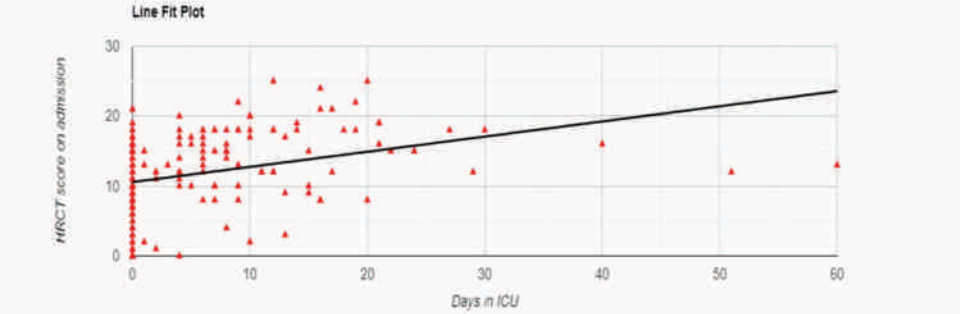


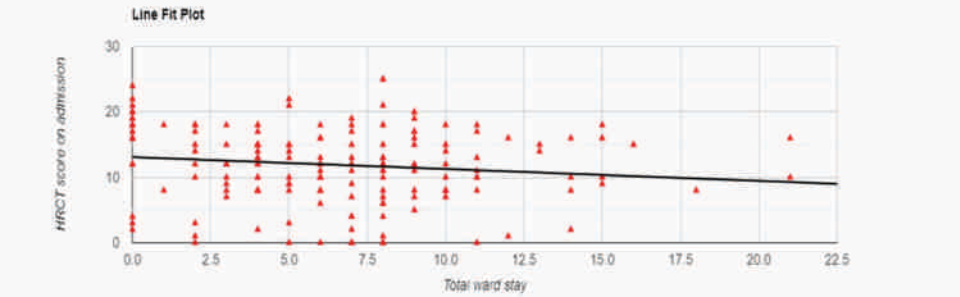
Figure 3: Pearson's correlation between days on oxygen and HRCT score on admission



Results of the Pearson correlation indicated that there is a **Significant** small positive relationship between Total Hospital stay and HRCT score on admission, ($r\ (165) = .288$, $p < .001$).



Results of the Pearson correlation indicated that there is a **Significant** medium positive relationship between Days in ICU and HRCT score on admission,($r\ (165) = .349$, $p < .001$).



Results of the Pearson correlation indicated that there is a ***Non-significant*** very small negative relationship between Total ward stay and HRCT score on admission, ($r\ (165) = .136$, $p = .080$).

Figure 4, 5 & 6 from above downwards

Table 4: Frequencies of Maximum Mode of Oxygen Support Required in Covid-19 patients

MODE OF OXYGENE SUPPORT	Total Number	Percentage
HFNC	4	2.4 %
HM	65	38.9 %
MV	16	9.6 %
NIV	30	18.0 %
NRBM	21	12.6 %
R/A	31	18.6 %

HFNC-High flow nasal cannula, HM- Hudson's mask, NIV- Non-invasive ventilation, NRBM- Non-rebreathing Mask, R/A- Room Air

The frequencies of the maximum mode of oxygen support required in Covid-19 patients in the present study showed that the most frequent maximum mode of oxygen support required was Hudson's face mask (HM) (38.9%) followed by non-invasive ventilation (NIV)(18.0%), non-rebreathing mask (NRBM) (12.6%), Mechanical ventilation (MV) (9.6%) and HFNC (2.4%), of patients while 18.6% of patients recovered without the requirement of oxygen.(table-4).

The cumulative percentage of patients requiring HFNC, HM, MV, NIV, NRBM, and R/A were 2.4%, 41.3%, 50.9%, 68.9%, 81.4%, and 100.0%, respectively. This suggests that a large proportion of patients required at least some form of oxygen support during their treatment.

Table 5: Outcome of COVID-19 patients in view of prevalence of comorbidities

Comorbidity	In Hospital Mortality	Discharged	Percent death
Hypertension (n=47)	7	40	14.89 %
Total Case of T2DM(n=63)	10	53	15.87 %
Known Case T2DM(n=41)	7	34	17.07 %
Newly Diagnosed T2DM (n=22)	3	19	13.64 %
CAD (n=6)	1	5	16.67 %
CVA (n=5)	0	5	0
Hyperthyroid(n=2)	2	0	100 %
Hypothyroid(n=10)	0	10	0
TB(n=1)	1	0	100 %
CKD(n=2)	1	1	50 %
AKI on CKD(n=2)	0	2	0
AKI(n=5)	3	2	60 %
Malignancy(n=3)	1	2	33.33 %
Non-comorbid	3	64	4.68%

The outcomes of 167 COVID-19 patients categorized based on their comorbidities showed that Diabetes Meletus had the highest number followed by hypertension. The number of other comorbid diseases was small (table-5).

HbA1C levels within that group.(table 6.1)

The study measured the levels of several inflammatory markers including C-reactive protein (CRP), D-dimer, erythrocyte sedimentation rate (ESR), ferritin, interleukin-6 (IL-6), lactate

Table 6: Relation of Covid-19 outcome in-view of comorbidities and their significance

	Discharged	**In-hospital mortality**
Non -Comorbid	**64**	**3**
Comorbid	**67**	**17**

The outcome of covid-19 patients in presence of comorbidities has brought a positive association between outcome and the presence of numbers of comorbidities (chi square test p-value <0.0485).further more there was positive association with outcome and the Type-2 diabetic patients with covid-19 infection (Chi square test p-value<0.030), but no association(p-value<0.0529) with hypertension (table-6)

In short, the observations suggest that comorbidities like hypertension, T2DM, CAD, and malignancy can increase the risk of death in COVID-19 patients. Hyperthyroidism and TB may also increase the risk of death, but due to the low number of cases it is difficult to comment. further research is needed. Hypothyroidism does not seem to increase the risk of death in COVID-19 patients. CKD and AKI can also be considered as risk factors, but AKI on CKD seems to have a lower death rate.(table 5 & 6)

In the present study out of a total of 167 covid-19 patients, 63 had associated Type-2 Diabetes out of which,22 were newly diagnosed, and the remaining 41 patients were a known cases of T2DM, with a mean HbA1C level of 8.89% and a standard deviation of 2.22.Both groups had relatively high mean HbA1C levels. However, the standard deviation was higher for the known case patients, indicating greater variability in

dehydrogenase (LDH), and procalcitonin (PCT). The levels of these markers were measured on admission and after 48 hours. on admission, levels of CRP, D-Dimer, ferritin, IL-6, LDH, and PCT were all significantly higher in patients with more severe disease compared to those with mild disease ($p<0.001$ for CRP, D-Dimer, IL-6, LDH, and PCT; $p=0.006$ for ferritin). After 48 hours, levels of CRP, D-Dimer, ferritin, IL-6, and PCT remained significantly higher in patients with more severe disease compared to those with mild disease ($p<0.05$ for CRP, D-Dimer, ferritin, and IL-6; $p<0.001$ for PCT), while levels of ESR did not differ significantly between the groups (table-7).

These findings suggest that levels of inflammatory markers may be useful in predicting disease severity in COVID-19 patients, particularly levels of CRP, D-Dimer, ferritin, IL-6, LDH, and PCT on admission. Monitoring changes in these markers over time may also provide important information about disease progression and treatment response.

Table 6.1: Comparison of HbA1C (percent) in cases of Type-2 Diabetes Mellitus

Case of T2DM (n=63)	**Number of Patients**	**Mean±SD**
Newly Diagnosed T2DM	22	8.88%+2.06
Known Case of T2DM	41	8.89%+2.22

Table 7: Comparison of Inflammatory Markers according to Clinical Disease Severity

Inflammatory Markers	Time Frame	Disease Severity			p-value
		Mild (n=42)	**Moderate (n=72)**	**Severe (n=53)**	
CRP (mg/dl)	On admission	19.5 (7.01-70.25)	41 (19-80.25)	77 (27-146.88)	**< .001**
	After 48 hours	10 (4-36)	18.5 (7-35)	28 (12-63)	0.240
D-dimer (ng/ml)	On admission	330 (220-720)	545 (250-1465)	1100 (260-2700)	**0.004**
	After 48 hours	250 (220-502.5)	485 (240-1245)	1340 (700-3140)	**< .001**
ESR ($mm 1^{st} hr$)	On admission	32.5 (26-41.75)	36.5 (27-52)	40 (31-66)	0.068
	After 48 hours	19 (13.25-28.5)	19 (14-29)	21 (14-32)	0.632
Ferritin(mcg/lit)	On admission	349.5 (207.25-575.75)	446.5 (330.75-671.25)	571 (340-955)	**0.006**
	After 48 hours	219.5 (105-317.5)	276 (173-446)	412 (210-800)	**< .001**
IL-6(pg/ml)	On admission	24.67 (7.46-39.85)	40.5 (16.18-74)	80 (24-176)	**< .001**
	After 48 hours	10.19 (4.81-21)	14.3 (6.13-32.5)	20.4 (8.87-70)	0.016
LDH (IU/lit)	On admission	299.5 (210-436.15)	402 (321.5-566.75)	472 (365-648)	**< .001**
	After 48 hours	202 (152.25-278)	258.5 (191.5-375)	377 (225-590)	**< .001**
PCT (%)	On admission	0.2 (0.1-0.36)	0.26 (0.18-0.39)	0.39 (0.25-0.48)	< .011
	After 48 hours	0.1 (0.02-0.17)	0.14 (0.05-0.24)	0.2 (0.1-0.36)	**< .001**
Values represent the Median and interquartile range					

CRP- C-reactive protein, ESR- Erythrocyte sedimentation rate, IL-6 – Interleukin-6,
LDH - Lactate dehydrogenase, PCT- Procalcitonin

Table 8: Comparison of Inflammatory Markers according to Outcome

INFLAMMATORY MARKERS	Time Frame	All patients (n=167)	Discharged (n=147)	Death (n=20)	p-value
CRP	On admission	45 (18.5-107)	43 (18.5-101.5)	53.35 (20.54-127)	0.464
	After 48 hours	19 (7-41)	16 (6.5-38.5)	26.5 (19-151)	**0.006**
D-DIMER	On admission	550 (234.5-1595)	490 (230-1245)	2350 (812.5-4000)	**< .001**
	After 48 hours	520 (240-1610)	450 (231.5-1220)	2925 (1262.5-4000)	**< .001**
ESR	On admission	37 (28-52.5)	35 (27-51.5)	41 (33.5-65.5)	0.081
	After 48 hours	19 (14-30.5)	19 (13-29)	29.5 (18.75-36.75)	0.060
FERRITIN	On admission	445 (300.8-722)	442 (273.5-671)	667.25 (328.5-1243.25)	**0.008**
	After 48 hours	285 (171-538.2)	270 (135-470.5)	462 (277-999)	**0.005**
IL-6	On admission	39 (16.62-89.15)	36 (16-71.5)	167.05 (78.7-330)	**< .001**
	After 48 hours	13.8 (6.75-34.26)	13 (6.15-29.5)	74 (19.75-112)	**< .001**
LDH	On admission	404 (307-564.5)	392 (301-549.5)	500.5 (416-836)	0.004
	After 48 hours	270 (190-391)	246 (186.5-360)	605 (424.5-702.5)	**< .001**
PCT	On admission	0.29 (0.18-0.4)	0.29 (0.18-0.41)	0.34 (0.2-0.39)	0.722
	After 48 hours	0.13 (0.08-0.28)	0.12 (0.06-0.25)	0.27 (0.1-0.48)	**0.03**
Values represent the Median and interquartile range					

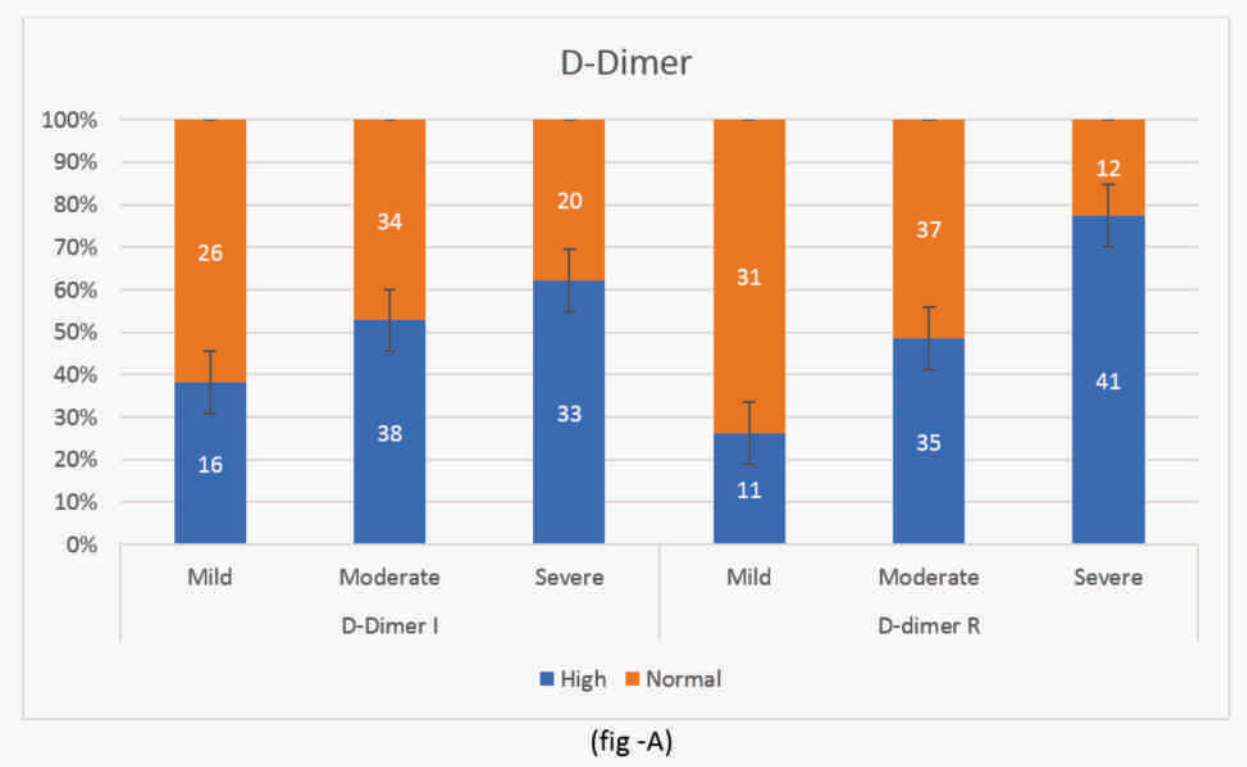
CRP- C-reactive protein, ESR- Erythrocyte sedimentation rate, IL-6 – Interleukin-6, LDH- Lactate dehydrogenase, PCT- Procalcitonin

On further analysis, it was observed that the levels of CRP, ESR, and ferritin were not significantly different between the discharged and deceased patients on admission. However, after 48 hours, the levels of CRP, ESR, and ferritin were significantly higher in the deceased group compared to the discharged group (table-8). In contrast, the levels of D-dimer, IL-6, and LDH were significantly higher ($p<0.001$) in the deceased group compared to the discharged group on admission and after 48 hours (fig 7,8 & 10)

The levels of PCT did not differ significantly between the two groups on admission, but after 48 hours, the levels of PCT were significantly higher($p=0.03$) in the deceased group compared to the discharged group (table-8).

In a nut-shell, this study suggests that higher levels of D-dimer, IL-6, and LDH are associated with poorer outcomes in COVID-19 patients. Additionally, increased levels of CRP, ESR, ferritin, and PCT after 48 hours may also be indicative of unfavourable prognosis.

Interestingly, out of 167 covid-19 patients selected 20 did not survive, 40 had severe symptoms and survived, and 107 had nonsevere symptoms.(table-9)



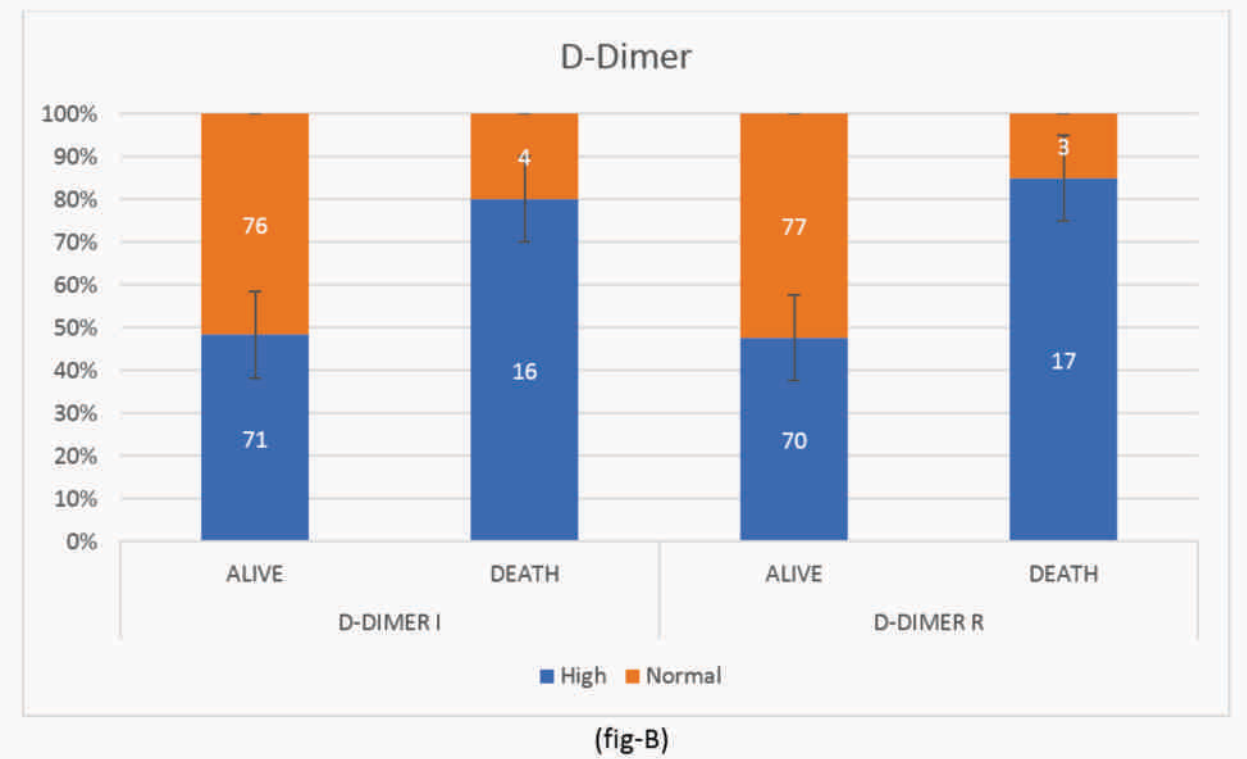
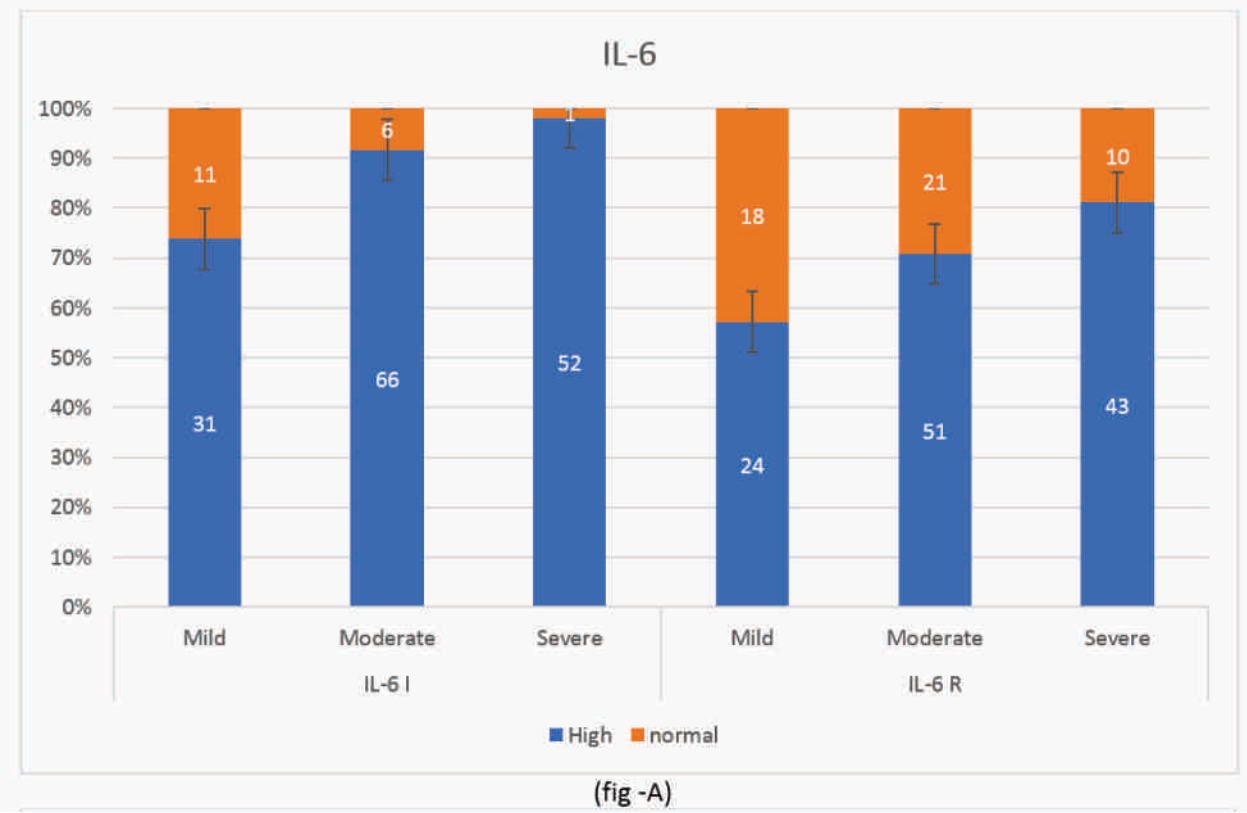


Figure 7: Comparison of a proportion of patients with elevated D-dimer levels on admission(D-dimer I) and after 48 hours(d-dimer R), based on clinical diseases severity (fig-A) and outcomes (fig-B).



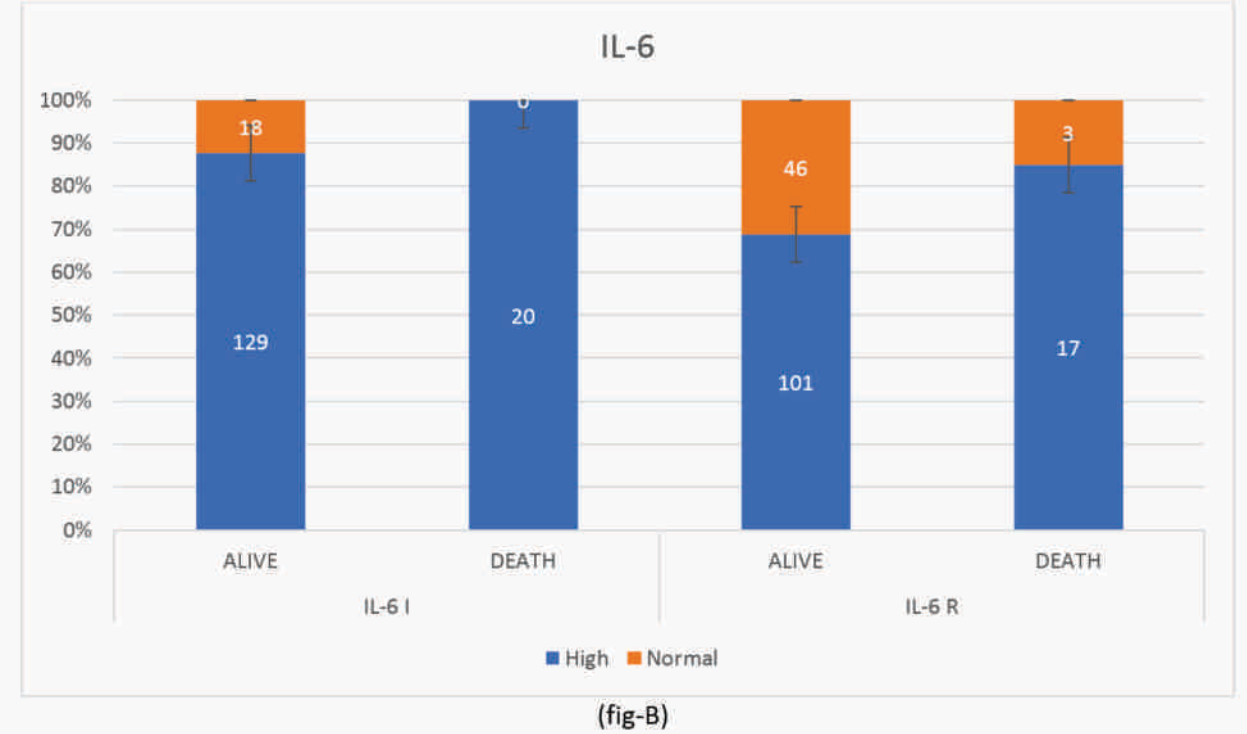
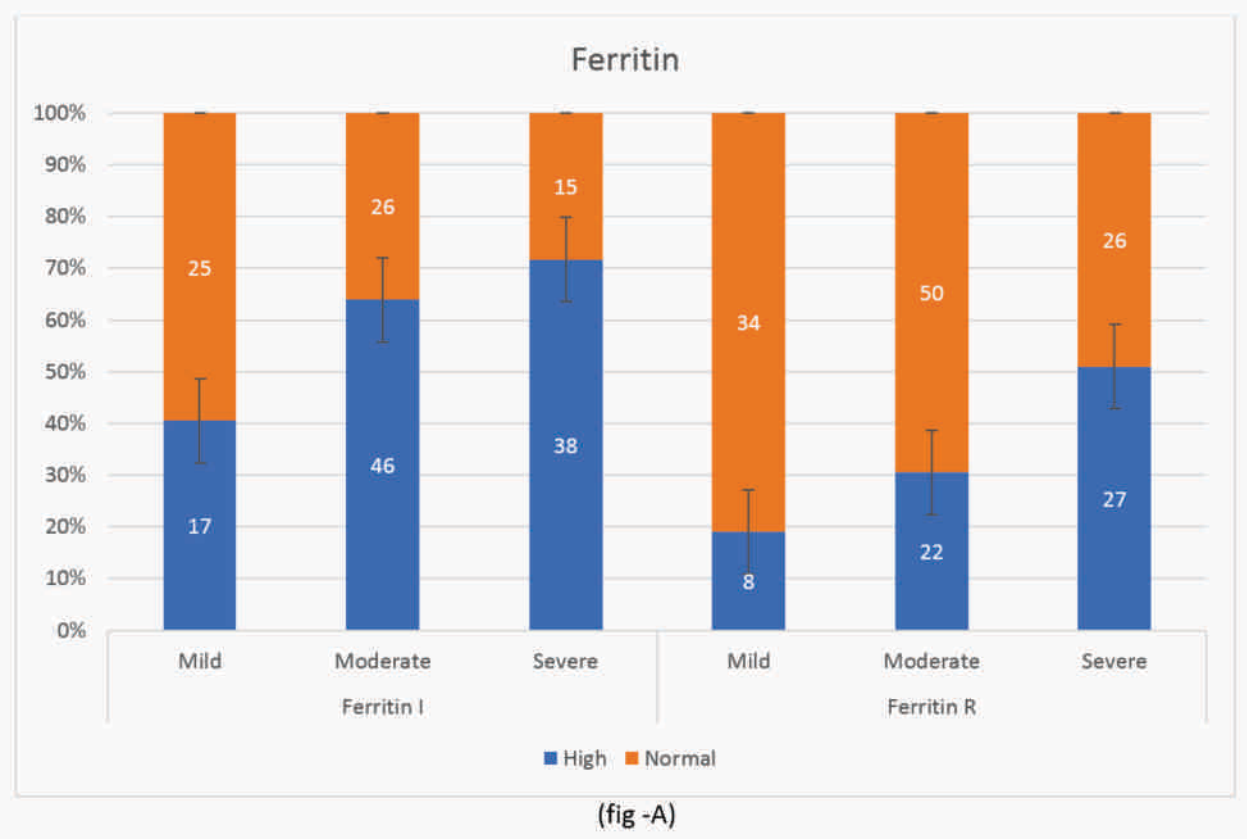
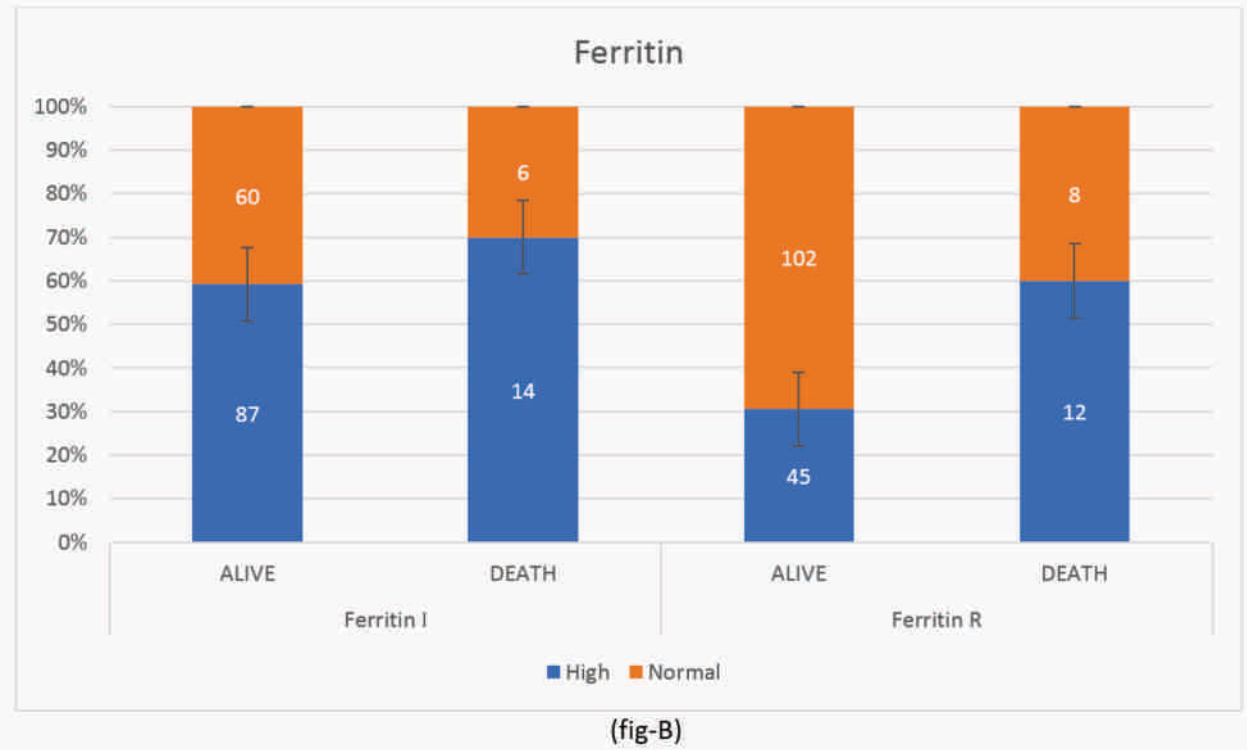


Figure 8: Comparison of a proportion of patients with elevated Serum IL-6 levels on admission (IL-6 I) and after 48 hours(IL-6 R), based on clinical diseases severity (fig-A) and outcomes (fig-B)

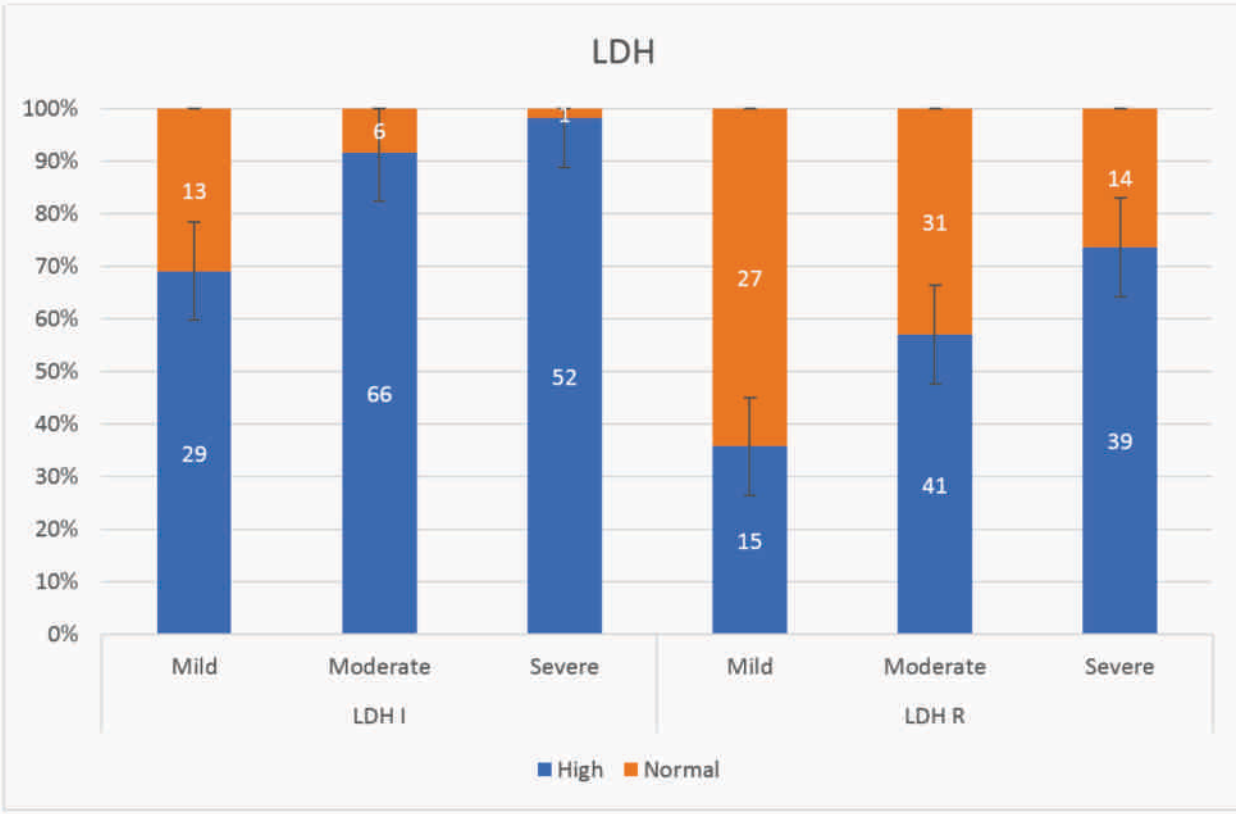


(fig -A)

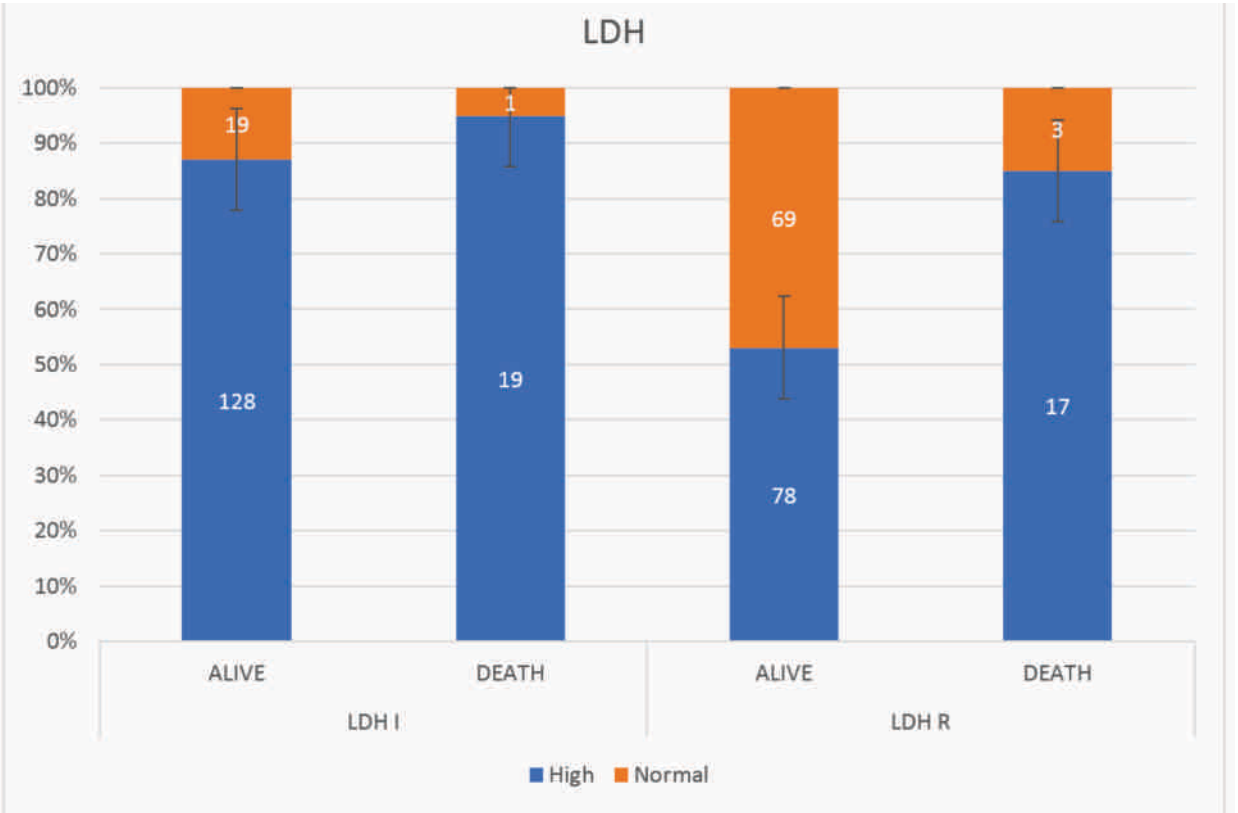


(fig-B)

Figure 9: Comparison of a proportion of patients with elevated Serum Ferritin levels on admission(Ferritin I) and after 48 hours(Ferritin R), based on clinical diseases severity (Fig-A) and outcomes (Fig-B)



(Fig -A)



(Fig-B)

Figure 10: Comparison of a proportion of patients with elevated Serum LDH levels on admission(LDH I) and after 48 hours(LDH R), based on clinical diseases severity (Fig-A) and outcomes (Fig-B)

Table 9: Demographics and baseline characteristics of Covid-19 patients according to Severity and Prognosis

		Disease severity and prognosis			p value		
Baseline characteristics	**All Patients (n=167)**	**(A) Nonsurvived (n=20)**	**(B) Severe & Survived (n=40)**	**(C) Nonsevere (n=107)**	**A vs B**	**B vs C**	**A vs C**
Age Median (IQR)	54 (42.5-64)	66 (56.3-72)	55.5 (44-64)	51 (39-62.5)	0.016	0.149	<0.001
Age Group							
<30 y	5	0	2	3	0.058	0.248	0.004
30-49 y	57	1	11	45			
50-69 y	105	19	27	59			
Gender							
Female	48	3	11	34	0.281	0.617	0.13
Male	119	17	29	73			
Co-morbidity							
Hypertension	47	7	13	27	0.846	0.378	0.365
Diabetes (Known case)	41	7	9	25	0.302	0.912	0.271
Diabetes (Newly diagnosed)	22	3	8	11	0.637	0.118	0.536
Renal dysfunction	9	4	0	5			
CAD	6	1	0	5			
CVA	5	0	2	3			
Hyperthyroid	2	2	0	0			
Hypothyroid	10	0	0	10			
Malignancy	3	1	0	2			
Other	15	4	5	6			

The median age of all patients was 54 years (IQR 42.5-64), Nonsurvived patients was 66 years (IQR 56.3-72), and of severe and survived patients was 55.5 years (IQR 44-64). The median age of Nonsevere patients was 51 years (IQR 39-62.5). The differences in age between Nonsurvived and severe and survived patients (p=0.016), Nonsurvived and Nonsevere patients (p<0.001), and severe and survived and Nonsevere patients (p=0.149) were statistically significant. However, the gender difference between different groups were not significant(table-9).

Evaluation of the presence of comorbidities in these patients showed that the most common comorbidity was hypertension, present in 47 patients. The differences in hypertension between Nonsurvived and severe and survived patients (p=0.846), Nonsurvived and Nonsevere patients (p=0.365), and severe and survived and Nonsevere patients (p=0.378) were not statistically significant. Likewise, 41 patients had diabetes as a known case, while 22 patients were newly diagnosed with diabetes. The differences in diabetic groups between nonsurvived and severe and survived patients (p=0.302), Nonsurvived and Nonsevere patients (p=0.271), and severe and survived and Nonsevere patients (p=0.912) were not statistically significant. Other comorbidities, such as renal dysfunction, CAD, CVA, hyperthyroidism, hypothyroidism, malignancy, and others, were also evaluated in this study (table-9).

Table 10: Inflammatory markers of Covid-19 patients according to Severity and Prognosis

		Disease severity and prognosis			p-value		
Inflammatory Markers	**Time Frame**	**A) Nonsurvived (n=20)**	**B) Severe & Survived (n=40)**	**C) Nonsevere (n=107)**	**A vs B**	**B vs C**	**A vs C**
CRP	On admission	53.35 (20.54-127)	79 (25.25-147.41)	39 (17.55-73.5)	0.393	**0.002**	0.178
	After 48 hours	26.5 (19-151)	27.6 (8.83-54.75)	13 (5-34)	0.153	0.046	0.002
D-dimer	On admission	2350 (812.5-4000)	705 (247.25-1887.5)	430 (230-1050)	**0.007**	**0.091**	**<.001**
	After 48 hours	2925 (1262.5-4000)	1050 (457.5-2892.5)	360 (220-860)	**0.022**	**<.001**	**<.001**
ESR	On admission	41 (33.5-65.5)	38 (30.75-63)	34 (27-49)	0.397	0.194	**0.047**
	After 48 hours	29.5 (18.75-36.75)	19.5 (12.75-31.25)	19 (13-29)	**0.034**	0.711	**0.006**
Ferritin	On admission	667.25 (328.5-1243.25)	546 (338.75-855.75)	428 (240.5-618)	0.41	**0.064**	**0.036**
	After 48 hours	462 (277-999)	383.5 (205-742.5)	221 (123-381)	0.236	**0.008**	**0.001**
IL-6	On admission	167.05 (78.7-330)	47.37 (21.52-146.75)	32 (12.6-54)	**0.013**	**0.01**	**<.001**
	After 48 hours	74 (19.75-112)	15.84 (8.38-33.49)	12 (5.05-26.43)	0.011	0.112	**<.001**
LDH	On admission	500.5 (416-836)	453 (369-645.75)	362 (277-503)	0.262	**0.001**	**<.001**
	After 48 hours	605 (424.5-702.5)	306 (219.75-432)	220 (178-311)	**0.001**	**0.001**	**<.001**
PCT	On admission	0.34 (0.2-0.39)	0.4 (0.25-0.53)	0.24 (0.15-0.39)	0.109	**<.001**	0.248
	After 48 hours	0.27 (0.1-0.48)	0.19 (0.1-0.32)	0.12 (0.04-0.21)	0.503	**0.002**	**0.009**
Values represent the Median and interquartile range							

CRP- C-reactive protein, ESR- Erythrocyte sedimentation rate, IL-6 – Interleukin-6, LDH- Lactate dehydrogenase, PCT- Procalcitonin

The present study also evaluated inflammatory markers of COVID-19 patients based on disease severity and prognosis (Table-10).

1. D-dimer

D-dimer levels in the study were significantly higher in nonsurvived and severe COVID-19 patients compared to Nonsevere patients on admission ($p<0.001$). After 48 hours, the D-dimer levels were still significantly higher in nonsurvived and severe COVID-19 patients compared to Nonsevere patients ($p<0.001$). Additionally, the D-dimer levels also remained significantly higher in severe COVID-19 patients who survived compared to Nonsevere patients ($p=0.022$). However, there was no significant difference between nonsurvived and severe COVID-19 patients in terms of D-dimer levels after 48 hours ($p=0.091$).

2. IL-6

The results showed that the median IL-6 level on admission was significantly higher in the Nonsurvived group (167 pg/mL) compared to the severe and survived group (47.37 pg/mL) and the Nonsevere group (32 pg/mL). The difference was statistically significant indicating that IL-6 may be a useful biomarker for predicting disease severity and prognosis in COVID-19 patients (table-10).

After 48 hours, the median IL-6 level decreased in all three groups. However, the decrease was more pronounced in the Nonsevere group (12 pg/mL) compared to the severe and survived group (15.84 pg/mL) and the non-survived group (74 pg/mL). The p-value for the comparison between the Nonsevere and nonsurvived groups was <0.001 suggesting that the decrease in IL-6 levels may be a sign of improvement in COVID-19 patients (table-10).

3.Serum Ferritin

There was a significant difference in ferritin levels between non-survivors and non-severe cases ($p=0.036$), with the non-survivors having higher levels (median: 445 ng/mL) than the non-severe cases (median: 667.25 ng/mL). The difference in ferritin levels between non-survivors and severe cases who survived was not statistically significant ($p=0.064$). There was also a significant difference in ferritin levels between severe cases who survived and non-severe cases ($p=0.036$), with the severe cases who survived having higher levels (median: 546 ng/mL) than the non-severe cases (median: 428 ng/mL). Moreover after 48 hours, there was a statistically significant decrease in all three groups (table-10).

4. LDH

On admission, the median LDH level in all patients was 404 (307-564.5) U/L. Nonsurvived patients had a higher median LDH level of 500.5 (416-836) U/L compared to severe and survived (453 (369-645.75) U/L) and Nonsevere (362 (277-503) U/L) patients, with a significant difference observed between A and B groups ($p=0.001$) and A and C groups ($p<0.001$). After 48 hours, the median LDH level in all patients decreased significantly. The results suggest that LDH levels are elevated in COVID-19 patients and may be a useful marker for disease severity and prognosis. Higher LDH levels on admission were associated with a higher risk of mortality, while higher LDH levels after 48 hours were associated with a higher risk of severe disease (Table-10).

5. CRP

CRP (C-reactive protein) is a marker of inflammation in the body the levels in COVID-19 patients have been found to be significantly elevated and are used as one of the indicators for the severity of the disease.

On admission, the CRP levels were highest in the nonsurvived group (53.35 mg/L), followed by the severe and survived group (79 mg/L), and the Nonsevere group (39 mg/L). However, the difference in CRP levels between the nonsurvived and severe and survived groups was not statistically significant ($p=0.393$), whereas the difference between the severe and survived and Nonsevere groups was significant ($p=0.002$).

After 48 hours, the CRP levels decreased in all groups, with the Nonsevere group showing the lowest levels (13 mg/L) and the severe and survived group showing the highest levels (27.6 mg/L). The difference in CRP levels between the nonsurvived and severe and survived groups was significant ($p=0.046$), whereas the differences between the nonsurvived and Nonsevere groups and the severe and survived and Nonsevere groups were not significant (Table-10).

Other laboratory parameters analyzed in the study include sodium (Na+), potassium (K+), neutrophil / lymphocyte ratio (NLR), and HbA1c. Na+ and K+ are electrolytes that play a critical role in maintaining the body's fluid and electrolyte balance. NLR is an indicator of the immune response, with high NLR values suggesting a more severe inflammatory response. HbA1c is a measure of long-term blood glucose control and is commonly used to diagnose and monitor diabetes.

The results showed that after 48 hours, the Na+ level was significantly higher in all patients compared to the levels on admission. However, there were no significant differences in Na+ levels between the different groups. K+ levels did not change significantly after 48 hours, and there were no significant differences between the patient groups. The NLR was significantly higher in the non survived group compared to the other groups on admission and after 48 hours. The NLR was also significantly higher in the severe and survived group compared to the Nonsevere group on admission, but not after 48 hours. The HbA1c level did not differ significantly between the patient groups on admission (Table-11).

The distribution of Covid-19 cases based on HRCT Score and

Table 11: Other Laboratory Parameters of Covid-19 patients according to Severity and Prognosis

			Disease severity and prognosis			p-value		
Parameters	**Time Frame**	**All Patients (n=167)**	**(A) Nonsurvived (n=20)**	**(B) Severe & Survived (n=40)**	**(C) Nonsevere (n=107)**	**A vs B**	**B vs C**	**A vs C**
Serum Sodium (NA^+)	On admission	134(129.5-138)	132.5 (128-137)	134.5 (130-137)	135 (130-138.5)	0.422	0.683	0.178
	After 48 hours	137(134-140)	134 (132.25-138.5)	138 (135.75-142)	136 (132-140)	**0.008**	**0.033**	0.239
Serum Potassium (K^+)	On admission	4.3 (4-4.7)	4.2 (3.68-5.05)	4.45 (4.08-4.7)	4.3 (4-4.7)	0.753	0.675	0.979
	After 48 hours	4.2 (4.1-4.8)	4.5 (4.1-4.83)	4.2 (3.98-4.6)	4.2 (4.1-4.8)	0.143	0.386	0.296
Neutrophil / Lymphocyte Ratio	On admission	5.333 (3.35-8.5)	10.88 (7.4-17.8)	5.63 (3.65-8)	4.56 (3.24-8)	**0.001**	0.34	**< .001**
	After 48 hours	5.333 (3.43-7.73)	7.97 (5.64-11.22)	4.85 (3.47-7.46)	4.88 (3.35-7.5)	**0.044**	0.67	**0.003**
HbA1C*	On admission	6.3 (5.85-7.9)	6.75 (5.98-9.43)	6.55 (6.1-8.18)	6.2 (5.8-7.65)	0.666	0.100	0.107

Values represent the Median and interquartile range

***No Significance of Repeat HbA1c**

Table 12: HRCT category vs Clinical category

Covid-19 category on the basis of HRCT Score	**Covid-19 category on the basis of Clinical Parameters**		
	Mild	Moderate	Severe
Mild (< 8)	24	19	5
Moderate (9 – 15)	18	31	25
Severe (>15)	0	22	23

Clinical Parameters were also analysed (Table-12). The rows indicate the Covid-19 category based on HRCT score, while the columns indicate the Covid-19 category based on clinical parameters. The values in the table represent the counts of patients falling into each category. The majority of patients fell into the moderate category based on clinical parameters, while the distribution of patients across the HRCT Score categories was more evenly spread. The highest count of patients is in the Moderate-Moderate category, indicating that a large proportion of patients have moderate Covid-19 based on both HRCT Score and clinical parameters. There was a smaller number of patients in the Severe-Severe category, indicating that fewer patients have severe Covid-19 based on both measures. Overall, the findings suggest that clinical parameters are a more sensitive indicator of Covid-19 severity than the HRCT Score.

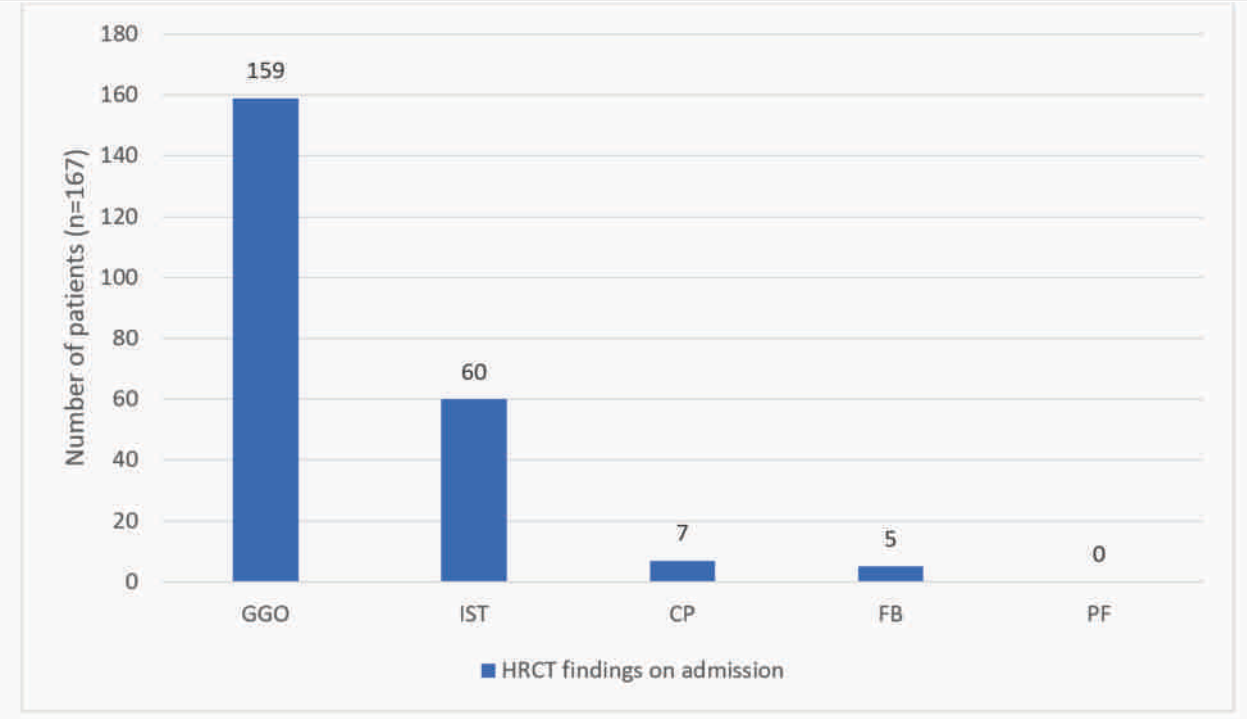


Figure 11: HRCT findings on Admission

GGO - Ground-glass opacities, IST- Interstitial thickening, CP- Crazy-paving patterns, FB- Fibrotic bands, PF-Pulmonary fibrosis

Analysis of HRCT findings on admission outrightly demonstrated that the majority of cases (95.2%) showed ground glass opacities (GGO) a characteristic finding in COVID-19 pneumonia. In addition to GGO, 60 patients (35.9%) had interstitial thickening (IST) and 7 patients (4.2%) had crazy-paving patterns (CP). Only 5 patients (3%) had fibrotic bands (FB) while none of the patients had established pulmonary fibrosis (PF) on their HRCT.

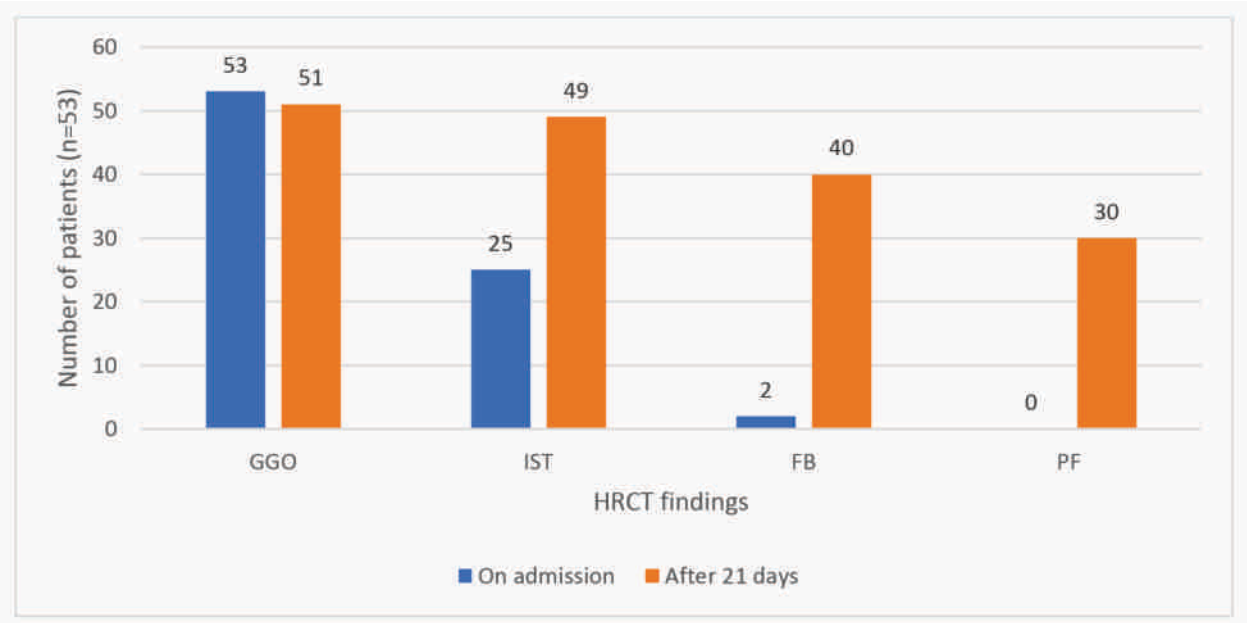


Figure 12: Comparison of HRCT findings on admission vs after 21 days

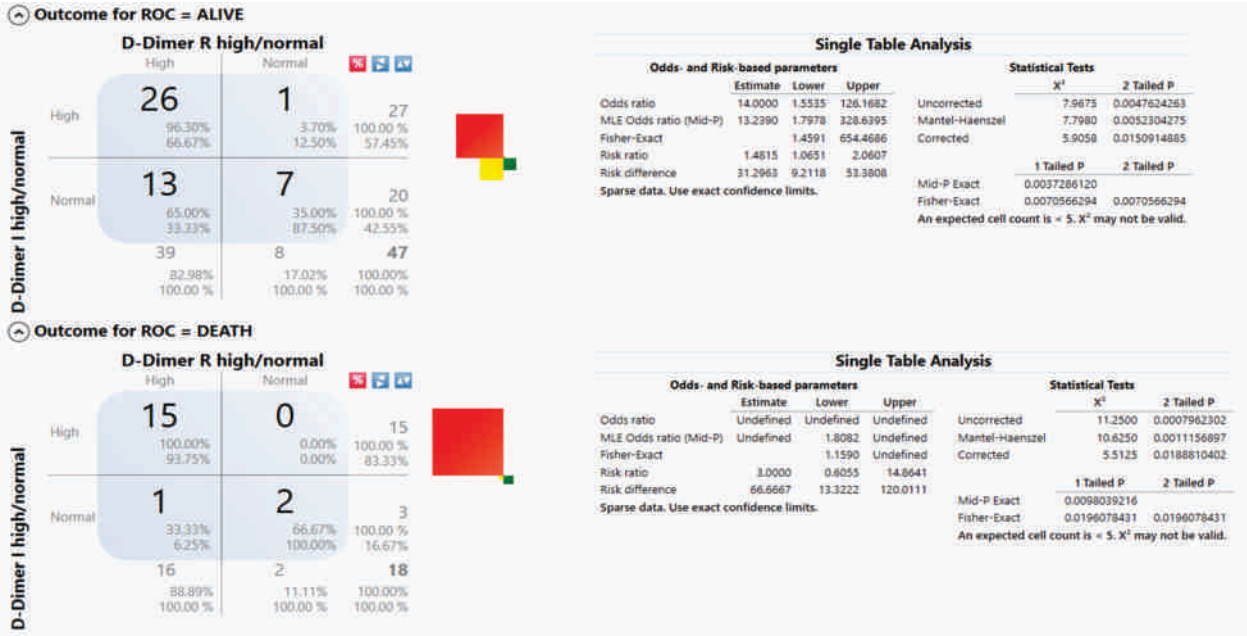
GGO - Ground-glass opacities, IST- Interstitial thickening, FB- Fibrotic bands, PF-Pulmonary fibrosis

Information based on the HRCT findings in patients with COVID-19, 53 patients required a repeat CT scan (Table-12) all 53 patients on admission showed ground-glass opacities (GGO) in their initial HRCT scans. Additionally, 25 patients showed interstitial thickening (IST), while only 2 patients had fibrotic bands (FB), and none of the patients had pulmonary fibrosis (PF) on their initial scans (Fig-12).

After 21 days, 51 patients still had GGO, indicating that their infection was still active, while 49 patients showed IST, suggesting that the inflammation in the lungs was still present in these patients. Notably, the number of patients with fibrotic bands increased from 2 to 40, suggesting that a significant proportion of patients were progressing toward pulmonary fibrosis. Finally, 30 patients showed pulmonary fibrosis on their repeat CT scans, indicating a more severe and chronic condition. Overall, these findings suggest that COVID-19 can cause significant lung damage and may progress to pulmonary fibrosis in some patients (Fig-12).

COVID-19 patients ICU on admission Progression and outcome

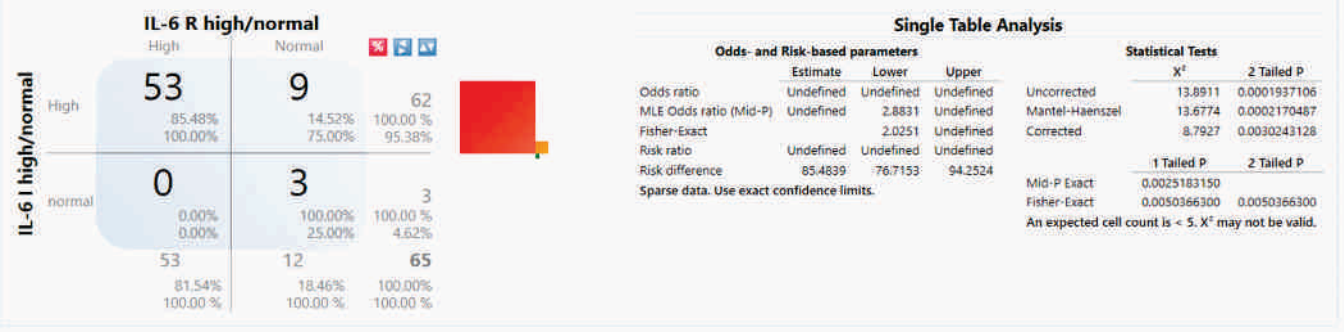
D-dimer is a fibrin degradation product that indicates the presence of blood clots. Covid-19 has been associated with an increased risk of blood clots, and high levels of D-dimer have been reported in severe cases. In this table, 42 patients had high D-dimer levels on admission, which increased to 55 after 48 hours. This suggests that the risk of blood clots may have increased in these patients over time.



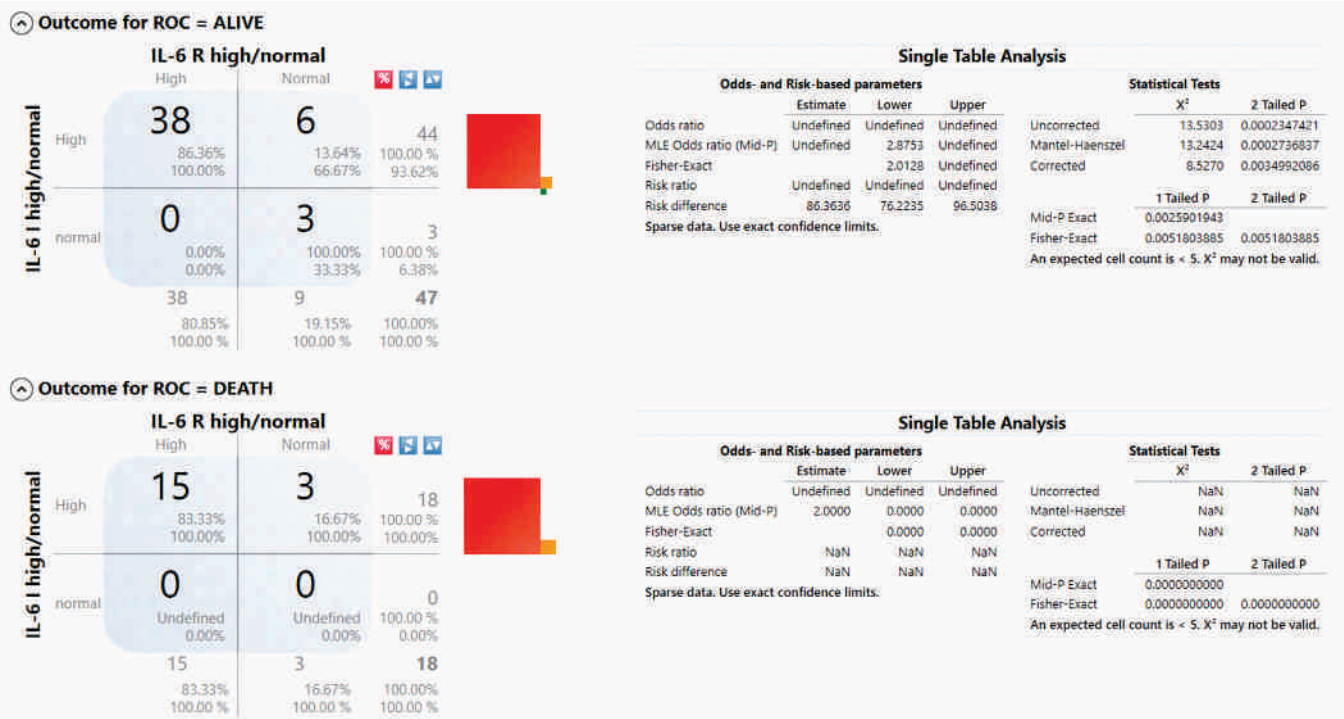
In this Graph, 15 out of 18 patients had high D-dimer levels on admission, which increased to 16 after 48 hours. This suggests that the risk of blood clots may have increased in these patients over time. However, the small sample size of the study limits the generalizability of these findings.

Figure 13: D-dimer on ICU admission and after 48 hours and the outcome

IL-6



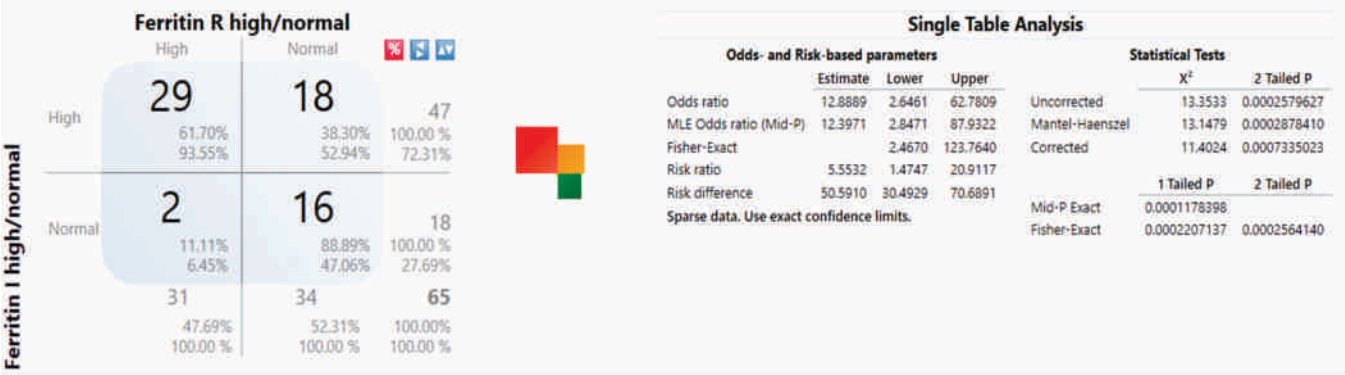
Interleukin-6 (IL-6) is a cytokine that plays a key role in the immune response. Elevated levels of IL-6 are associated with systemic inflammation and are often seen in patients with severe Covid-19. In this table, 62 patients had high levels of IL-6 on admission, which decreased to 53 after 48 hours. This suggests that the patients' immune response may have improved over time, potentially due to treatment or a natural improvement in their condition but not significant improvement.



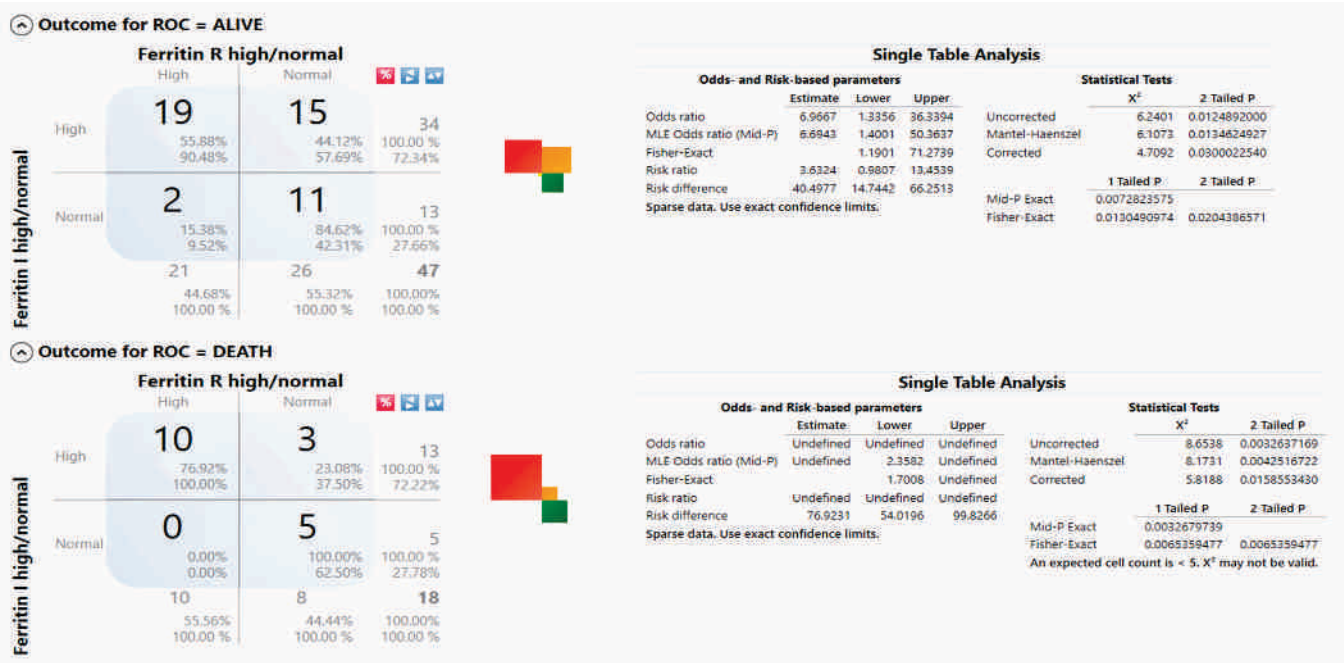
In this Graph, all 18 patients had high levels of IL-6 on admission, which decreased to 15 after 48 hours. This suggests that the patients' immune response may have improved slightly over time, potentially due to treatment or a natural improvement in their condition.

Figure 14: Interleukin-6 (IL-6) on ICU admission and after 48 hours and the outcome

S.Ferritn



Ferritin is a protein that stores iron in the body. Elevated levels of ferritin are often seen in patients with inflammation, infection, or malignancy. In patients with Covid-19, high levels of ferritin have been associated with a poor prognosis. In this table, 47 patients had high ferritin levels on admission, which decreased to 31 after 48 hours. This suggests that the patients' inflammation may have decreased over time, potentially due to treatment or a natural improvement in their condition.

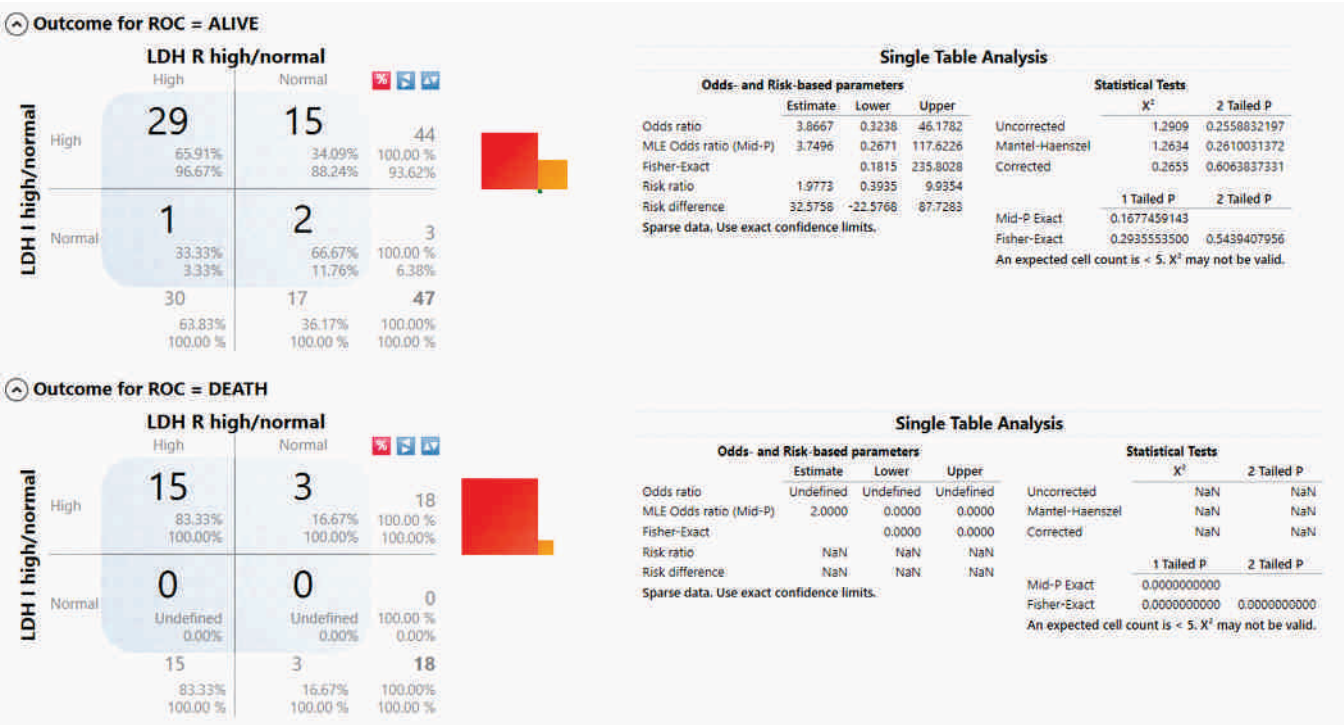


In patients with Covid-19, high levels of ferritin have been associated with a poor prognosis. In this table, 13 out of 18 patients had high ferritin levels on admission, which decreased to 10 after 48 hours. This suggests that the patients' inflammation may have decreased slightly over time, potentially due to treatment or a natural improvement in their condition.

Figure 15: S.ferritin on ICU admission and after 48 hours and the outcome

LDH

LDH (lactate dehydrogenase) is an enzyme that is released when cells are damaged or destroyed. Elevated levels of LDH are often seen in patients with tissue damage, such as in acute respiratory distress syndrome (ARDS) which can be a complication of Covid-19. In this table, 62 patients had high levels of LDH on admission, which decreased to 45 after 48 hours. This suggests that the patients' tissue damage may have improved over time, potentially due to treatment or a natural improvement in their condition.



In this table, all 18 patients had high levels of LDH on admission, which decreased to 15 after 48 hours. This suggests that the patients' tissue damage may have improved slightly over time, potentially due to treatment or a natural improvement in their condition.

Figure 16: Lactate Dehydrogenase (LDH) on ICU admission and after 48 hours and the outcome

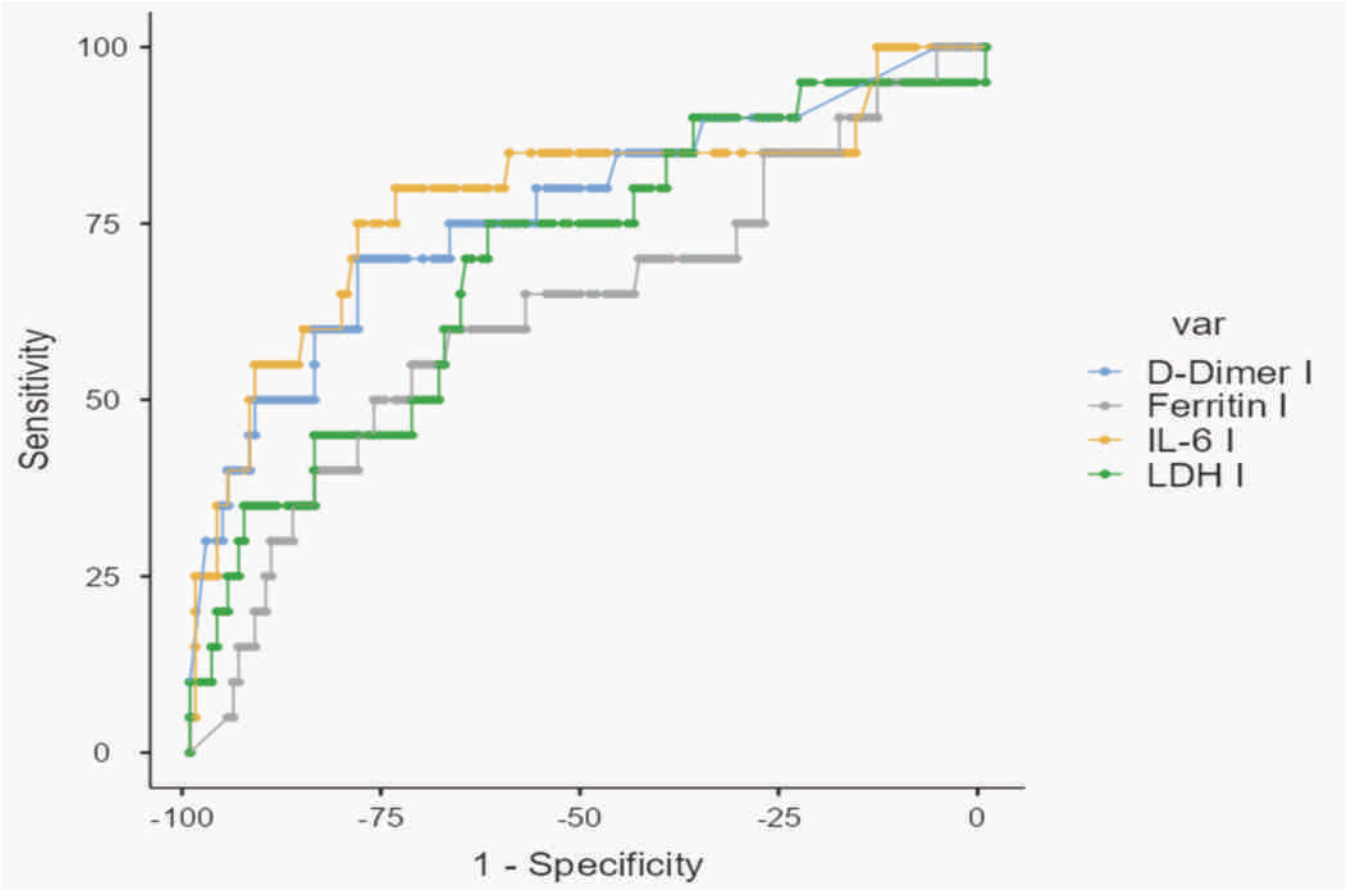


Figure 17: 17 ROC Curve of Inflammatory Markers as a Predictor of Covid-19 Mortality

Overall, the evaluation of inflammatory markers is an important tool for monitoring and managing Covid-19 patients in the ICU. It can provide valuable information on the patients' immune response, inflammation, and tissue damage, and help guide treatment decisions. However, it is important to interpret these markers in the context of other clinical factors and use them as part of a comprehensive approach to patient care.

The role of inflammatory markers in predicting COVID-19 mortality was evaluated using the ROC curve. Survivors were considered negative and severe patients as positive. The area under the ROC curve (ROC-AUC) was assessed (Figure-17).

Table 13: Inflammatory Markers on admission and their respective Pearson Correlation Coefficients with HRCT lung Involvement

Inflammatory Markers	**Correlation Coefficients**
IL-6	0.275974088
D-Dimer	0.067948852
S.Ferritin	0.238913546
LDH	0.287165217
CRP	0.179717074

CRP- C-reactive protein, IL-6 – Interleukin-6, LDH- lactate dehydrogenase

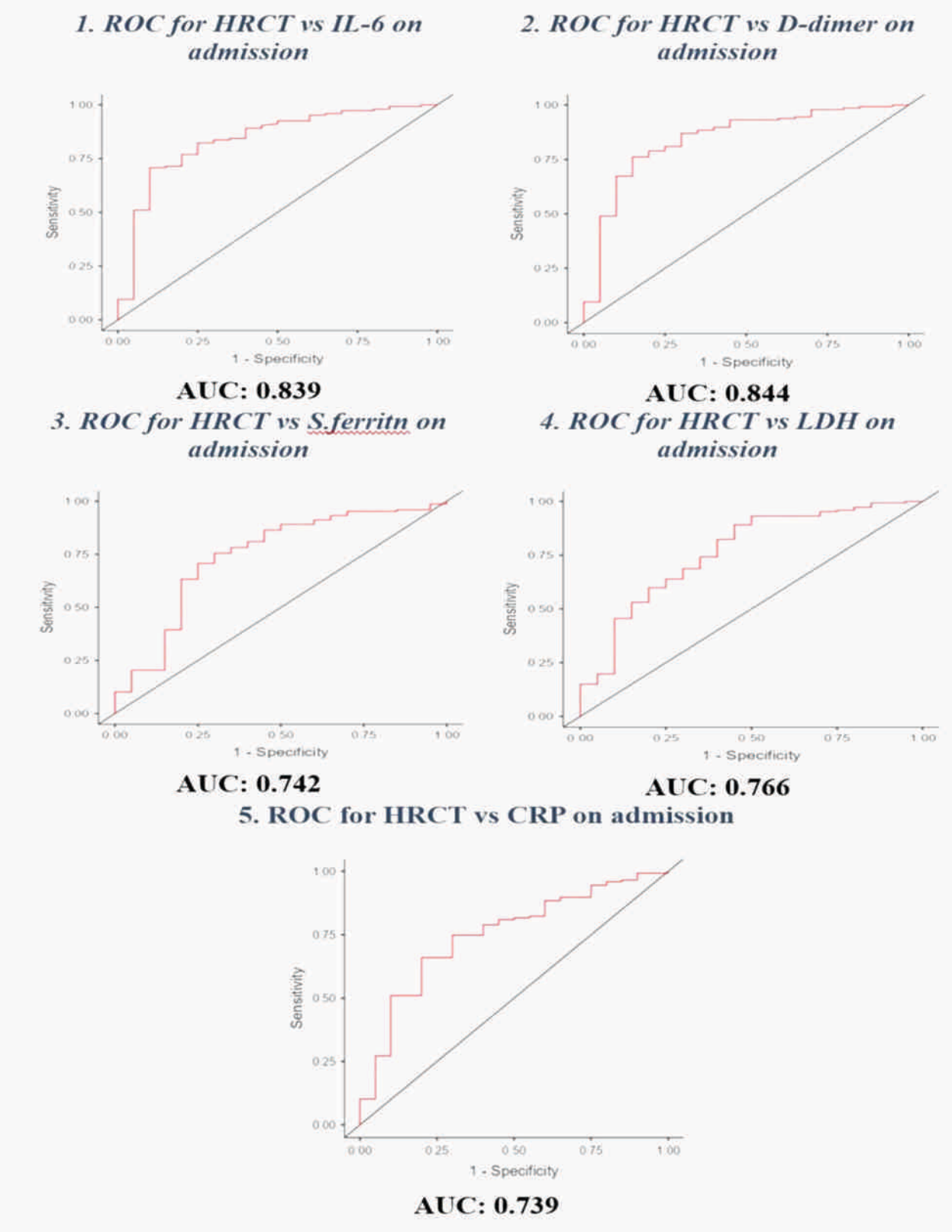


Figure 18: ROC curves for various inflammatory markers vs HRCT severity. HRCT cut-offs were taken to be ,25%, 25-49%, 50-74%, =. 75% lung involvement. (AUC : Area under curve)

DISCUSSION

COVID -19 virus took over the reins of healthcare system in a flash. The world immediately was quickly battling a pandemic coveted by the virus. The organism in itself along with the illness is still a mystery waiting to be solved. We must understand the disorder at the grassroot level, maybe try and identify the fundamental question which still lingers each and every healthcare professional – "What does the virus do to our body exactly?"

The available tools for us to evaluate disease severity for COVID-19 include the basic radiological evaluation comprising of HRCT Scan thorax and laboratory parameters in form of inflammatory markers (Mainly IL-6, D-Dimer, CRP, Ferritin, LDH, N/L Ratio and ESR). We tried to outline and characterise disease severity and progression via laboratory parameters and extrapolating the data on presenting radiological presentation and clinical profile. Universally disease has been termed as Mild Moderate and Severe according to Lung involvement in a form of numeric score.

- HRCT Severity Index 1-8 Mild Disease
- HRCT Severity Index 9-15 Moderate Disease
- HRCT Severity Index 16-25 Severe Disease.

But what was observed in the present study was that the raw score strictly based on lung involvement only did not reflect the severity and predict the progression of disease severity accurately. Laboratory parameters had more statistical significance than radiological parameters.

- **AGE**

Age has always been a critical risk factor, not just in primary infection but also for vulnerability towards getting infected, disease severity, disease progression and mortalities. Chen Y *et al*[46] observed that it is unmistakable that aging is an important risk factor for severe COVID-19 disease and its adverse health outcomes including hospitalization, ICU admission, and death. The study mentions the hypothesis that with age the physiological reserves of Immunosenescence are depleted and older individuals are more at risk for severe COVID-19 and mortality. The median age in our study was more than 50 years, and all the patients who had or progressed towards a severe disease were older individuals, relatable with the findings of universal consensus.

- **GENDER**

COVID-19 has not been significantly gender biased, specifically in terms of prevalence, but outcome tells a different story. The one-of-a-kind study done by Jin *et al*[47]. investigated the role of gender in morbidity and mortality in COVID -19 and concluded that men were at more risk for worse outcomes and death, independently of age. The more plausible reasoning can be weaker immunity due to genetic and hormonal factors[48], or lifestyle differences[49] or normal serum testosterone level facilitating systemic dissemination preceded by viral entry[50], Males also have a higher number of ACE-2 receptors in their body which has higher affinity to bind with SARS CoV-2. Our findings resound the same observation, not just the severe cases but overall, the demographic distribution had male preponderance[50].

- **Comorbidities**

COVID-19 infections favoured individuals with pre-existing comorbidities much more. It can be understood from the concepts of immune system, that comorbidities, especially poorly controlled illness had a major role in principle infection, rapid disease progression and failure to respond to adequate therapy[51]. But interestingly in our population co-morbidities had no correlation with disease progression or mortality. Highest number patients were diabetic, and all of them had poor glycaemic control (Mean HbA1C 8.8%). In new onset diabetes the mean HbA1C was 8.89%. but interestingly it had no impact on disease. However this sheds light on a different problem our nation is facing altogether that is of Diabetes awareness, follow up and treatment compliance.

Clinically COVID-19 exhibits a quite familiar picture with SARS illness, due to its similarity with the previously infective strains seen in MERS. The most common symptoms are cough and fever, as observed worldwide. However, the patients who had severe diseases tend to have hypoxia and ultimately shortness of breath as a more profound manifestations masking the other symptoms[51]. Even in the current setting, patients who were dyspnoeic or breathing normally on room air had strong significance ($p < 0.001$) in terms of disease outcome.

- **Hypoxia and O_2 Requirment**

Hypoxia in COVID -19 had significant impact on severity and is associated with poor outcomes[52]. Hypoxia is a condition where the body or a part of it does not receive enough oxygen. In COVID-19, hypoxia can be caused by the following mechanisms[53-58]:

a. Impaired blood flow and oxygenation in the lungs due to inflammation, clotting and damage to the blood vessels

b. Reduced ability of red blood cells to carry oxygen due to viral infection or immune response.

c. Dysfunctional response of the body to low oxygen levels, such as lack of hypoxic vasoconstriction (narrowing of blood vessels in low-oxygen areas) or impaired chemoreceptors (sensors that detect changes in blood oxygen levels).

Some COVID-19 patients may experience "happy hypoxia" where they have very low blood oxygen levels but do not feel any symptoms or discomfort. This can be dangerous as it may delay seeking medical attention and increase the risk of organ damage[58].

• The severe the hypoxia and longer the duration of O_2 requirement the higher the mortality. This part was observed in a study done on 140 patients of Moderate to Critical COVID - 19 requiring oxygen supplementation in China[59]. They observed that Patients with SpO_2 values of 90% or less were older and were more likely to be men, to have hypertension, and to present with dyspnoea than those with SpO2 values greater than 90%. Overall, 36 patients (25.7%) died during hospitalization after median 14 - day follow-up. Higher SpO_2 levels after oxygen supplementation were associated with reduced mortality independently of age and sex (hazard ratio per 1-U SpO_2, 0.93; 95% CI, 0.91 to 0.95; P<.001). This led to a conclusion that hypoxia is independently associated with in–hospital mortality. Critically ill patients with COVID-19, SpO2 values greater than 90% with oxygen supplementation indicate a very high likelihood of survival. These patients should, thus, receive maximal supportive care during the acute illness. Participants in our study had a decreasing chance of survival with increasing days of O_2 requirement. Patient's on admission while dividing in mild, moderate and severe category the Mean Hypoxia and range of SpO_2 [Mild- 95.71 (1.49), Moderate- 93.81 (2.46), Severe- 83.58 (8.26)] was in declining order which further during in hospital evaluation gives idea that it is directly proportional to the Mean duration of O_2 requirement[Mild- 4.67 (7.89), Moderate- 8.31 (8.06), Severe -14.08 (12.13) in days], which suggests has high positive significance(p-value < 0.001) of hypoxia on admission with diseases severity and outcome.

• **HRCT**

HRCT remained the diagnostic tool of choice for all the healthcare professionals for quick assessment of disease extent and possibility of future progression. However, the caveat may be, world still believed that CT scan gives a raw idea of the disease state and future progression. Length of stay in hospital and ICU was directly proportional to the presenting finding and severity score of HRCTs, also noted by Goyal *et al*[60].

• **Inflammatory Markers**

COVID-19 disease ultimately is an inflammatory reaction towards a foreign invasion. Hence, we cannot emphasize the role of inflammatory biomarkers enough. Understanding the role of inflammatory cytokines, chemokines, and clinical immunology will be the approach to find out the possible novel therapeutic interventions. Therapies involving regulation of immune responses help in inhibiting the various steps in the pathologies of infection. Also, updated knowledge regarding the dysregulation of immune system and disease outcome in critically ill patients serves as a precautionary measure in the development and evaluation of vaccine. In this prospective analysis we have studied the principle inflammatory cytokines and their trend throughout the disease progression. They include CRP, IL-6, D-Dimer, LDH, Ferritin and Neutrophil / Lymphocyte (N/L) ratio.

Levels of **CRP** had an increasing trend in proportion to increasing disease severity. But this finding was unrelated to the outcome. The only way CRP had an impact on outcome was a refractory increase after 48 hours of the initial test. This has been consistent with other studies[61,62] In a study by Mueller et al.63 Obsereved that, patients requiring ICU-level care at any point during their hospital course had elevated CRP, D-dimer, procalcitonin, and IL-6 levels, compared to patients who remained on the non-ICU medical floor throughout their hospitalization. CRP can be a strong predictor of disease severity, a helpful marker for active inflammation and can help guide pulsed steroid therapy and tapering at any given time.

LDH is an active marker of cell damage and injury. Commonly used for cardiac muscle and cell status and anaemia, it is also an important inflammatory marker. However, its usefulness in attesting damage extent of COVID-19 disease at cellular level is still in dilemma. Vijaykumari *et al*[64]. evaluated role of various inflammatory markers including LDH. In that study LDH exerted a significant association with clinical severity whereas on admission as well as on day seven, LDH showed significant association with radiological severity, LDH done on day one, day five, and day seven showed significant association with outcome in COVID-19 in our study population. Szarpak L *et al.*[65] reported a mean LDH of 154.49 U/L in COVID-19 and observed LDH as a COVID-19 severity marker and predictor for survival whereas in our study mean LDH was noted as 336.03 ± 327.55 U/L. We report that LDH is strongly significant for disease outcome [p-value (on admission 0.04) and (after 48 hours <0.001)]. Along with that raised LDH after 48 hours had a p-value of 0.003 for disease progression towards severity and had even stronger impact (p <0.001) for the outcome. Along with IL-6, LDH was indicating a severe disease and leading to poor outcome in ICU patients as in our study all 18 patients who died were admitted in ICU on admission had high levels of IL-6 and LDH and were not that much improved after 48 hours evaluation unlike the same case with other inflammatory markers. This can be explained by understanding both of their pathophysiology together and establishing link between them as IL-6 and LDH are both involved in the hyperinflammatory response that leads to severe outcomes in COVID-19 patients[66-69]. IL-6 is a cytokine that regulates immune cells and inflammation[69]. LDH is an enzyme that catalyses the conversion of lactate to pyruvate[66].

When COVID-19 infects the lungs, it triggers an excessive production of IL-6 that causes a cytokine storm[69]. This leads to tissue damage, hypoxia, organ failure and ARDS[68-69]. LDH levels reflect the extent of tissue damage and hypoxia caused by COVID-19[66]. High levels of IL-6 and LDH are associated with increased risk of respiratory failure and death[66-68].

IL-6 levels trigger the CRP synthesis in hepatic cells has been at the forefront of COVID-19 infection. Grifoni *et al*[70]. studied IL-6 as a prognosticator in patients with COVID-19. The study commented on the available evidence that purposed pro-inflammatory cytokines in a pivotal role in the

pathophysiology of COVID induced lung damage. They stated that Interleukin-6 (IL-6) is one of the main mediators of inflammatory and immune response initiated by infection or injury and increased levels of IL-6 are found in more than one half of patients with COVID-19[71].

Levels of IL-6 seem to be associated with inflammatory response, respiratory failure, needing for mechanical ventilation and/or intubation and mortality in COVID-19 patients. In a meta-analysis including nine studies (total 1426 patients) reporting on IL-6 and outcome in COVID-19, mean IL-6 levels were more than three times higher in patients with complicated COVID-19 compared with those with non-complicated disease, and IL-6 levels were associated with mortality risk[79]. This importance of IL-6 resonates the same findings in our study. What we found that deranged IL-6 was the strongest in outlining disease progression and had the highest impact on outcome of the patient. IL-6 along with D-Dimer and Ferritin, done at any point of time during the course of the illness gave the most accurate picture of the disease state and future predictability. In a cohort analysis of IL-6 levels showed a positive correlation with CRP in patients who had IL-6 levels drawn, and patients treated with tocilizumab, an IL-6 receptor monoclonal antibody, had rapid and sustained decrease in CRP levels. The analysis suggested that increased CRP rise, and by virtue presumed elevation of IL-6, in the first 24–48 h may be of critical importance to disease progression; no other study is focused only on this hyper-acute period[63]. In early prediction of a potential requirement of mechanical ventilation, Herold *et al.*[80] concluded that the maximal level of IL-6, followed by CRP level, was highly predictive of the need for mechanical ventilation. This suggests the possibility of using IL-6 or CRP level to guide escalation of treatment in patients with COVID-19–related hyperinflammatory syndrome.

Ferritin is a protein that stores iron and can be used as a cheap and reliable indicator of inflammation in COVID-19 pandemic. Ferritin has been well studied in bacterial infection, and it can help to evaluate COVID-19 pneumonia patients before starting indoor treatment compared to other markers and CT severity. Ferritin levels are very important for predicting how severe COVID-19 pneumonia is; especially, changes in ferritin levels can guide interventions in critical care settings. It is also important to check ferritin levels with other factors like how long the patient has been sick, how well they can breathe, and when they need BIPAP/NIV support. Ferritin levels can show if COVID pneumonia is getting worse, as some patients with mild CT findings and normal initial ferritin levels have developed critical conditions, and we have seen that rising ferritin levels along with other markers can sometimes mean bacterial infection in the second week of illness and require specific therapy. Ferritin levels can also help to estimate the risk of lung fibrosis after COVID pneumonia and monitor lung inflammation in suspected cases of fibrosis with this easy-to-use marker[81]. In a study of 1000 COVID -19 cases significant association with ferritin in prediction severity of COVID-19 was found[81]. Dahan *et al*[82]. observed role of ferritin as "severity predictor" in their study with raised ferritin level in advanced CT severity cases. Ferritin remained equally significant in our study also. At any point of time during the illness Ferritin was significant in relation to the final outcome. On admission raised ferritin correctly predicted hypoxia. Only place ferritin was not helpful was in predicting mortality as it had negative relation in disease progression towards severe to death.

D-Dimer variability was seen on initial evaluation, more significantly between discharged and fatal patients. However, multivariate regression analysis did not identify D-Dimer as an independent risk factor of the mortality in severe COVID-19 patients. This has been consistently reported in other studies for example Chen *et al*[83]. However, in stark contrast D-Dimer has been associated with poor prognosis also[84]. High level of D-dimer is indicative of developing thrombosis. A recent study on COVID-19 patients in Wuhan revealed an extremely high incidence of thrombosis (41/48, 85.4%) in severe patients with a death rate of 31.7% (13/41)85. The talk on importance of D-Dimer is highly individualized and variable. But given the thromboembolic nature of the COVID-19 in mind, D-Dimer undeniably remains an unmissable marker. What we did observe in our study is at any given moment in COVID-19 D-Dimer is the most significant marker (on admission p-value <0.004, after 48 hours p-value <0.001 for all the groups) for disease progression. It remained the strongest predictor of disease outcome (on admission p-value <0.001, after 48 hours p-value <0.001).

- **Neutrophil / Lymphocyte Ratio (NLR)**

Higher Neutrophil / Lymphocyte Ratio (NLR) is suggested to be another independent risk factor in mortality in COVID-19[86]. The talked about analysis took cohorts of patients with COVID-19 with a baseline NLR and exposed the data to a univariate and multivariate logistic regression model. The authors concluded that there was 8% higher risk of in-hospital mortality for each unit increase in NLR. NLR is an independent risk factor of the in-hospital mortality for COVID-19 patients especially for male. Assessment of NLR may help identify high risk individuals with COVID-19. Qin *et al*[87]. studied 452 patients with COVID-19 and saw that severe cases tend to have an abnormally high level of NLR. Our study derived NLR as a strongly significant marker for disease severity and outcome.

- **Procalcitonin (PCT)**

PCT, which is the 116-amino acid precursor of the hormone calcitonin, is normally synthesised and released by thyroid parafollicular C cells. However, it can also be synthesised in many extra thyroid tissues during bacterial infection, which is mediated by increased concentrations of tumour necrosis factor-alpha (TNFα) and interleukin 6[88]. A meta-analysis also demonstrated that increased PCT values are related to an ~5-fold higher risk of severe COVID-19[89]. A study by Hu R *et al*[90].

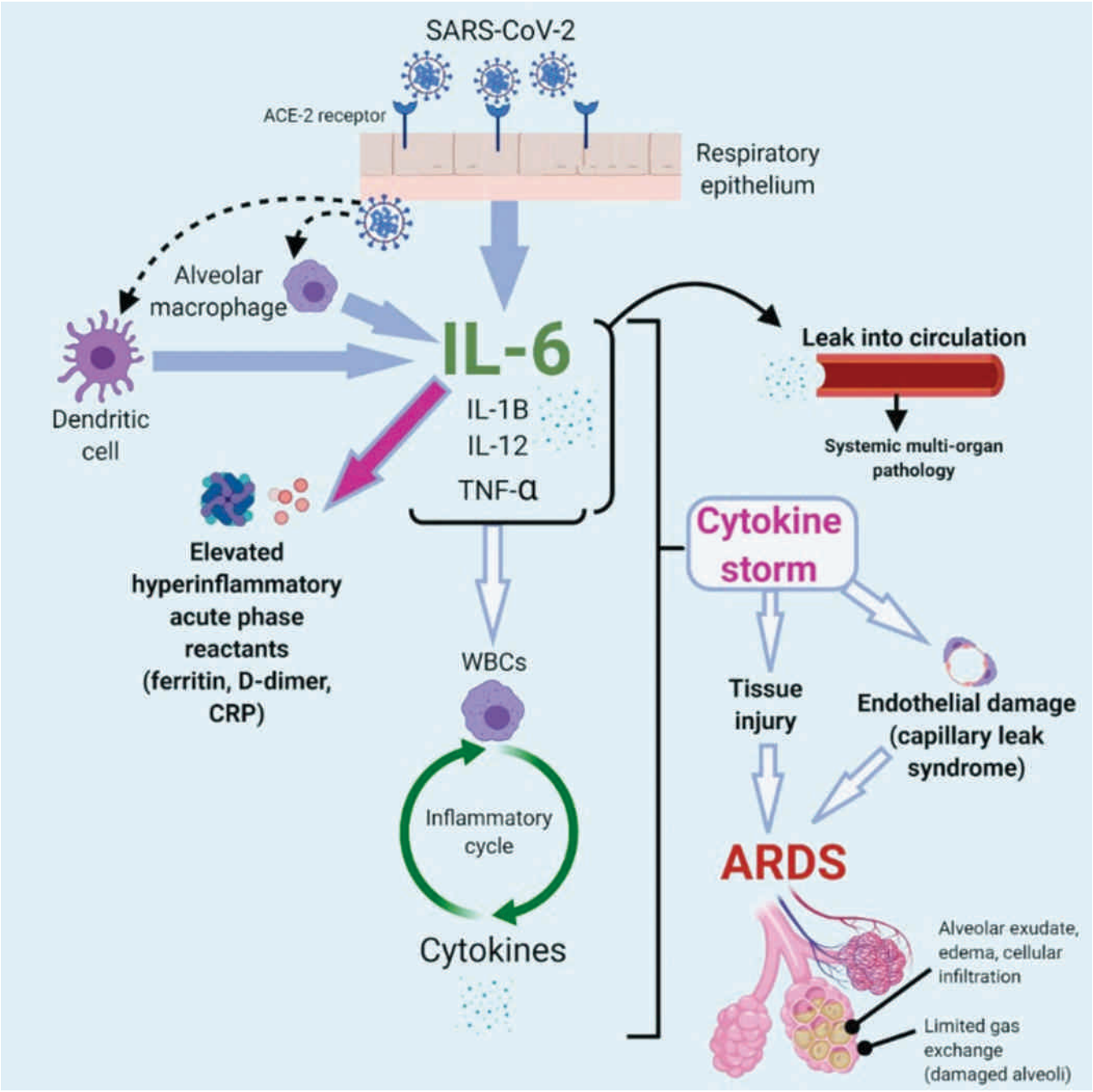


Figure 19: Proposed model of the cytokine storm in severe COVID-19, revealing the mechanistic complexity of the cytokine cascade and subsequent pathology[72-77]

The model is primarily based on current knowledge of cytokine storm, as seen in macrophage activation syndrome (MAS), hemophagocytic lymphohistiocytosis (HLH), and chimeric antigen receptor (CAR) T-cell therapy. Notably, cytokine storm in both MAS and HLH displays increased levels of cytokines interleukin (IL)-1, IL-2, IL-18, macrophage colony stimulating factor (M-CSF), interferon-γ, and tumor necrosis factor (TNF) α, in addition to IL-6, although definitive causality due to all these cytokines has not been determined[78].

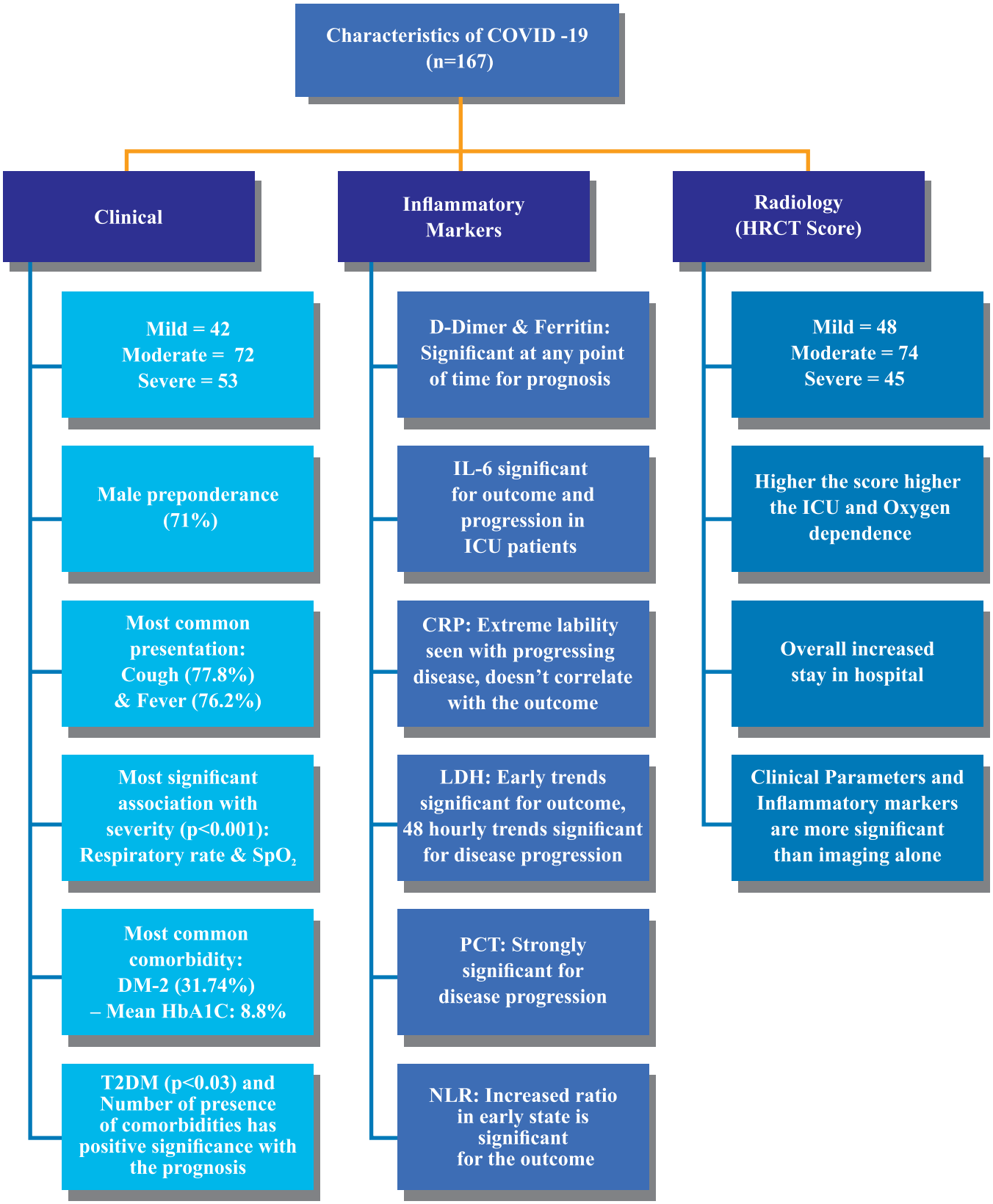
evaluated 95 patients retrospectively and observed that total serum PCT levels increased exponentially as the disease progressed towards severe manifestation. They concluded that PCT may be an indicator of disease severity and may contribute to determining the severity of patients with COVID-19. In addition, serial PCT measurements may be useful in predicting the prognosis. Additional investigation is needed to further illustrate the mechanisms by which increased PCT is synthesised and released in patients infected with SARS-CoV-2. Our study had a p-value of 0.002 of PCT's correlation with disease progression and even stronger association (p 0.009) with disease outcome.

- **Serum Electrolytes**

Syndrome of Inappropriate Anti -Diuretic Hormone can occur in the evolution of inflammatory diseases of infectious or non-infectious causes, malignant diseases, cardiovascular or hepatic diseases, but also in the evolution of acute respiratory distress syndrome (ARDS)[91,92] Ho and colleagues reported the first case of COVID-19 associated with SIADH manifested by new-onset seizures[93]. Yousaf *et al.* published a series of cases of patients diagnosed with COVID-19 who also associated SIADH[94] All patients included in this study had severe acute hyponatremia. After excluding other possible aetiologies, these authors established that this hydro electrolytic disorder is secondary to SIADH. Commonly ignored and almost always inadequately treated, Hyponatraemia can be a major cause of refractory illness and can potentially lead to irreversible end term cranial complications. Majority of patients in our study were hyponatraemic and remained so even during the time of discharge / death. Hence, hyponatraemia had a strong significance on disease severity, more so after 48 hours of initial evaluation. Early diagnosis, meticulous search for aetiology and efficient management of salt and water, could help a favourable outcome even for a severe COVID-19 illness. Interestingly the most logical counterpart potassium did not alter the disease course or modify the disease outcome in any tests performed.

The most frustrating aspect of COVID-19 undeniably remains it's multifactorial causation and multisystemic involvement. Target organ can be lungs, but the infection encompasses entire physiological process of homeostasis. In return, the immune system has a fairly dysregulated host response, which is proving to be frugal than adequate. However strong the odds may be, it is always stacked against you in terms of catching the dreaded infection, as the world has seen it already. However less the mortality percentage may be, more infection always means more deaths. We cannot ignore the number of deaths by just looking at the percentage of maths. In searching for answers, it does seem disheartening that we are left with more and more questions in the end, but more and more quality research will build a solid foundation for a robust understanding of the virus and carve out an effective plan to tackle it, than what we are having at the present moment in form of vaccines and supportive measures.

STUDY SUMMARY



CONFLICT OF INTEREST: None

FINANCIAL SUPPORT: None

REFERENCES

1. Zhu N, Zhang D, Wang W, Li X, Yang B, Song J, *et al.* A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med. 2020 Feb 20;382(8):727–33.
2. The Novel Coronavirus Pneumonia Emergency Response Epidemiology Team. The Epidemiological Characteristics of an Outbreak of 2019 Novel Coronavirus Diseases (COVID-19) - China, 2020. China CDC Wkly. 2020 Feb 21;2(8):113–22.
3. Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, *et al.* Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. Lancet. 2020 Feb 15;395(10223):507–13.
4. China: WHO Coronavirus Disease (COVID-19) Dashboard With Vaccination Data | WHO Coronavirus (COVID-19) Dashboard With Vaccination Data [Internet]. [cited 2022 Dec 15]. Available from: https://covid19.who.int/region/wpro/country/cn
5. WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020 [Internet]. [cited 2022 Dec 15]. Available from: https://www.who.int/director-general/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020
6. MoHFW | Home [Internet]. [cited 2022 Dec 15]. Available from: https://www.mohfw.gov.in/
7. WHO Coronavirus (COVID-19) Dashboard | WHO Coronavirus (COVID-19) Dashboard With Vaccination Data [Internet]. [cited 2022 Dec 15]. Available from: https://covid19.who.int/
8. India: WHO Coronavirus Disease (COVID-19) Dashboard With Vaccination Data | WHO Coronavirus (COVID-19) Dashboard With Vaccination Data [Internet]. [cited 2022 Dec 15]. Available from: https://covid19.who.int/region/searo/country/in
9. Bhandari S, Bhargava A, Sharma S, Keshwani P, Sharma R, Banerjee S. Clinical Profile of Covid-19 Infected Patients Admitted in a Tertiary Care Hospital in North India. J Assoc Physicians India. 2020 May;68(5):13–7.
10. Number of Cases [Internet]. [cited 2022 Dec 16]. Available from: https://prsindia.org/covid-19/cases
11. Belser JA, Rota PA, Tumpey TM. Ocular tropism of respiratory viruses. Microbiol Mol Biol Rev. 2013 Mar;77(1):144–56.
12. Guan W jie, Ni Z yi, Hu Y, Liang W hua, Ou C quan, He J xing, *et al.* Clinical Characteristics of Coronavirus Disease 2019 in China. New England Journal of Medicine. 2020 Apr 30;382(18):1708–20.
13. Dariya B, Nagaraju GP. Understanding novel COVID-19: Its impact on organ failure and risk assessment for diabetic and cancer patients. Cytokine Growth Factor Rev. 2020 Jun;53:43–52.
14. ACE2 protein expression summary - The Human Protein Atlas [Internet]. [cited 2022 Dec 16]. Available from: https://www.proteinatlas.org/ENSG00000130234-ACE2
15. Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, Transmission, Diagnosis, and Treatment of Coronavirus Disease 2019 (COVID-19): A Review. JAMA. 2020 Aug 25;324(8):782–93.
16. Dong X, Cao YY, Lu XX, Zhang JJ, Du H, Yan YQ, *et al.* Eleven faces of coronavirus disease 2019. Allergy. 2020 Jul;75(7):1699–709.
17. Wu C, Chen X, Cai Y, Xia J, Zhou X, Xu S, *et al.* Risk Factors Associated With Acute Respiratory Distress Syndrome and Death in Patients With Coronavirus Disease 2019 Pneumonia in Wuhan, China. JAMA Intern Med. 2020 Jul 1;180(7):934.
18. Yang X, Yu Y, Xu J, Shu H, Xia J, Liu H, *et al.* Clinical course and outcomes of critically ill patients with SARS-CoV-2 pneumonia in Wuhan, China: a single-centered, retrospective, observational study. Lancet Respir Med. 2020 May;8(5):475–81.
19. Li X, Xu S, Yu M, Wang K, Tao Y, Zhou Y, *et al.* Risk factors for severity and mortality in adult COVID-19 inpatients in Wuhan. Journal of Allergy and Clinical Immunology. 2020 Jul;146(1):110–8.
20. Wang K, Zhang Z, Yu M, Tao Y, Xie M. 15-day mortality and associated risk factors for hospitalized patients with COVID-19 in Wuhan, China: an ambispective observational cohort study. Intensive Care Med. 2020 Jul 23;46(7):1472–4.
21. Liu Y, Sun W, Guo Y, Chen L, Zhang L, Zhao S, *et al.* Association between platelet parameters and mortality in coronavirus disease 2019: Retrospective cohort study. Platelets. 2020 May 18;31(4):490–6.
22. Ponti G, Maccaferri M, Ruini C, Tomasi A, Ozben T. Biomarkers associated with COVID-19 disease progression. Crit Rev Clin Lab Sci. 2020 Aug 17;57(6):389–99.
23. Yu HH, Qin C, Chen M, Wang W, Tian DS. D-dimer level is associated with the severity of COVID-19. Thromb Res. 2020 Nov;195:219–25.

24.Tan C, Huang Y, Shi F, Tan K, Ma Q, Chen Y, *et al.* C-reactive protein correlates with computed tomographic findings and predicts severe COVID-19 early. J Med Virol. 2020 Jul 25;92(7):856–62.

25.Henry BM, Aggarwal G, Wong J, Benoit S, Vikse J, Plebani M, *et al.* Lactate dehydrogenase levels predict coronavirus disease 2019 (COVID-19) severity and mortality: A pooled analysis. Am J Emerg Med. 2020 Sep;38(9):1722–6.

26.Zhang C, Wu Z, Li JW, Zhao H, Wang GQ. Cytokine release syndrome in severe COVID-19: interleukin-6 receptor antagonist tocilizumab may be the key to reduce mortality. Int J Antimicrob Agents. 2020 May;55(5):105954.

27.Han H, Ma Q, Li C, Liu R, Zhao L, Wang W, et al. Profiling serum cytokines in COVID-19 patients reveals IL-6 and IL-10 are disease severity predictors. Emerg Microbes Infect. 2020 Jan 1;9(1):1123–30.

28.Cooper ID, Crofts CAP, DiNicolantonio JJ, Malhotra A, Elliott B, Kyriakidou Y, et al. Relationships between hyperinsulinaemia, magnesium, vitamin D, thrombosis and COVID-19: rationale for clinical management. Open Heart. 2020 Sep 16;7(2):e001356.

29.Sverzellati N, Milanese G, Milone F, Balbi M, Ledda RE, Silva M. Integrated Radiologic Algorithm for COVID-19 Pandemic. J Thorac Imaging. 2020 Jul;35(4):228–33.

30.COVID-19 Updates | RANZCR [Internet]. [cited 2023 Jan 15]. Available from: https://www.ranzcr.com/our-work/coronavirus

31.Coronavirus (COVID-19) clinical radiology resources | The Royal College of Radiologists [Internet]. [cited 2023 Jan 15]. Available from: https://www.rcr.ac.uk/college/coronavirus-covid-19-what-rcr-doing/coronavirus-covid-19-clinical-radiology-resources

32.Dennie C, Hague C, Lim RS, Manos D, Memauri BF, Nguyen ET, *et al.* Canadian Society of Thoracic Radiology/Canadian Association of Radiologists Consensus Statement Regarding Chest Imaging in Suspected and Confirmed COVID-19. Canadian Association of Radiologists Journal. 2020 Nov 1;71(4):470–81.

33.Li K, Fang Y, Li W, Pan C, Qin P, Zhong Y, *et al.* CT image visual quantitative evaluation and clinical classification of coronavirus disease (COVID-19). Eur Radiol. 2020 Aug 25;30(8):4407–16.

34.Huang L, Han R, Ai T, Yu P, Kang H, Tao Q, *et al.* Serial Quantitative Chest CT Assessment of COVID-19: A Deep Learning Approach. Radiol Cardiothorac Imaging. 2020 Apr 1;2(2):e200075.

35.Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, *et al.* Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. The Lancet. 2020 Feb;395(10223):497–506.

36.Lei J, Li J, Li X, Qi X. CT Imaging of the 2019 Novel Coronavirus (2019-nCoV) Pneumonia. Radiology. 2020 Apr;295(1):18–18.

37.Nishiyama A, Kawata N, Yokota H, Sugiura T, Matsumura Y, Higashide T, *et al.* A predictive factor for patients with acute respiratory distress syndrome: CT lung volumetry of the well-aerated region as an automated method. Eur J Radiol. 2020 Jan;122:108748.

38.Gattinoni L, Caironi P, Cressoni M, Chiumello D, Ranieri VM, Quintel M, *et al.* Lung Recruitment in Patients with the Acute Respiratory Distress Syndrome. New England Journal of Medicine. 2006 Apr 27;354(17):1775–86.

39.Mossa-Basha M, Meltzer CC, Kim DC, Tuite MJ, Kolli KP, Tan BS. Radiology Department Preparedness for COVID-19: *Radiology* Scientific Expert Review Panel. Radiology. 2020 Aug;296(2):E106–12.

40.Chung M, Bernheim A, Mei X, Zhang N, Huang M, Zeng X, *et al.* CT Imaging Features of 2019 Novel Coronavirus (2019-nCoV). Radiology. 2020 Apr;295(1):202–7.

41.Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation [Internet]. [cited 2023 Jan 16]. Available from: https://www.science.org/doi/epdf/10.1126/science.abb2507?src=getftr

42.Udugama B, Kadhiresan P, Kozlowski HN, Malekjahani A, Osborne M, Li VYC, et al. Diagnosing COVID-19: The Disease and Tools for Detection. ACS Nano. 2020 Apr 28;14(4):3822–35.

43.Praharaj I, Jain A, Singh M, Balakrishnan A, Dhodapkar R, Borkakoty B, *et al.* Pooled testing for COVID-19 diagnosis by real-time RT-PCR: A multi-site comparative evaluation of 5- & 10-sample pooling. Indian Journal of Medical Research. 2020;152(1):88.

44.Wiersinga WJ, Rhodes A, Cheng AC, Peacock SJ, Prescott HC. Pathophysiology, Transmission, Diagnosis, and Treatment of Coronavirus Disease 2019 (COVID-19): A Review. JAMA [Internet]. 2020 Aug 25 [cited 2023 Feb 26];324(8):782–93. Available from: https://jamanetwork.com/journals/jama/fullarticle/2768391

45.jamovi - open statistical software for the desktop and cloud [Internet]. [cited 2023 Feb 26]. Available from: https://www.jamovi.org/

46.Chen Y, Klein SL, Garibaldi BT, Li H, Wu C, Osevala NM, *et al.* Aging in COVID-19: Vulnerability, immunity and intervention. Ageing Res Rev. 2021 Jan;65:101205.

47.Jin JM, Bai P, He W, Wu F, Liu XF, Han DM, *et al.* Gender Differences in Patients With COVID-19: Focus on Severity and Mortality. Front Public Health. 2020;8:152.

48.Gao Y, Ding M, Dong X, Zhang J, Kursat Azkur A, Azkur D, *et al.* Risk factors for severe and critically ill COVID-19 patients: A review. Allergy. 2021 Feb 4;76(2):428–55.

49.Papadopoulos V, Li L, Samplaski M. Why does COVID-19 kill more elderly men than women? Is there a role for testosterone? Andrology. 2021 Jan 5;9(1):65–72.

50.Shaw AC, Joshi S, Greenwood H, Panda A, Lord JM. Aging of the innate immune system. Curr Opin Immunol. 2010 Aug;22(4):507–13.

51.Hentsch L, Cocetta S, Allali G, Santana I, Eason R, Adam E, *et al.* Breathlessness and COVID-19: A Call for Research. Respiration. 2021;100(10):1016–26.

52.Kashani KB. Hypoxia in COVID-19: Sign of Severity or Cause for Poor Outcomes. Mayo Clin Proc. 2020 Jun;95(6):1094–6.

53.Chan ED, Sharma V. Does Hypoxia Itself Beget Worsening Hypoxemia in COVID-19? Mayo Clin Proc [Internet]. 2021 Mar 1 [cited 2023 Feb 28];96(3):824–5. Available from: http://www.mayoclinicproceedings.org/article/S0025619621000434/fulltext

54.What Causes Hypoxemia in COVID-19? (Part I) - Mass General Advances in Motion [Internet]. [cited 2023 Feb 28]. Available from: https://advances.massgeneral.org/research-and-innovation/article.aspx?id=1313

55.Dhont S, Derom E, van Braeckel E, Depuydt P, Lambrecht BN. The pathophysiology of "happy" hypoxemia in COVID-19. Respir Res [Internet]. 2020 Jul 28 [cited 2023 Feb 28];21(1). Available from: https://pubmed.ncbi.nlm.nih.gov/32723327/

56.Nitsure M, Sarangi B, Shankar GH, Reddy VS, Walimbe A, Sharma V, *et al.* Mechanisms of Hypoxia in COVID-19 Patients: A Pathophysiologic Reflection. Indian J Crit Care Med [Internet]. 2020 [cited 2023 Feb 28];24(10):967–70. Available from: https://pubmed.ncbi.nlm.nih.gov/33281323/

57.Bohn MK, Hall A, Sepiashvili L, Jung B, Steele S, Adeli K. Pathophysiology of COVID-19: Mechanisms underlying disease severity and progression. Physiology [Internet]. 2020 Sep 1 [cited 2023 Feb 28];35(5):288–301. Available from: https://journals.physiology.org/doi/10.1152/physiol.00019.2020

58.Happy Hypoxia: COVID-19 May Cause Dangerously Low Oxygen Levels With Few Other Symptoms [Internet]. [cited 2023 Feb 28]. Available from: https://www.verywellhealth.com/covid-19-happy-hypoxia-oxygen-levels-5090027

59.Xie J, Covassin N, Fan Z, Singh P, Gao W, Li G, *et al.* Association Between Hypoxemia and Mortality in Patients With COVID-19. Mayo Clin Proc. 2020 Jun;95(6):1138–47.

60.Goyal A, Antani M, Agarwal S, Gadara C, Shah M, Bhatt CJ. Correlation of the severity of HRCT findings with the other hematologic parameters & viral load in patients with confirmed COVID – 19 Infection. Asian J Med Sci. 2021 Sep 1;12(9):4–10.

61.Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, *et al.* Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet [Internet]. 2020 Mar 28 [cited 2023 Feb 25];395(10229):1054–62. Available from: https://pubmed.ncbi.nlm.nih.gov/32171076/

62.Ruan Q, Yang K, Wang W, Jiang L, Song J. Clinical predictors of mortality due to COVID-19 based on an analysis of data of 150 patients from Wuhan, China. Intensive Care Med [Internet]. 2020 May 1 [cited 2023 Feb 25];46(5):846. Available from: /pmc/articles/PMC7080116/

63.Mueller AA, Tamura T, Crowley CP, DeGrado JR, Haider H, Jezmir JL, *et al.* Inflammatory Biomarker Trends Predict Respiratory Decline in COVID-19 Patients. Cell Rep Med. 2020 Nov;1(8):100144.

64.Vijayakumari V, Mayilananthi K, Reddy SP, Kumar K, Vijayakumari V, Krishnan D. Role of Inflammatory Markers in Predicting Severity and Outcome in COVID-19 Patients Attending a Tertiary Care Institute of Tamil Nadu. Journal of Communicable Diseases (E-ISSN: 2581-351X & P-ISSN: 0019-5138) [Internet]. 2022 Mar 16 [cited 2023 Feb 25];2022:186–94. Available from: https://medical.advancedresearchpublications.com/index.php/Journal-CommunicableDiseases/article/view/818

65.Szarpak L, Ruetzler K, Safiejko K, Hampel M, Pruc M, Kanczuga - Koda L, *et al.* Lactate dehydrogenase level as a COVID-19 severity marker. Am J Emerg Med [Internet]. 2021 Jul 1 [cited 2023 Feb 25];45:638–9. Available from: https://pubmed.ncbi.nlm.nih.gov/33246860/

66.Liu T, Zhang J, Yang Y, Ma H, Li Z, Zhang J, et al. The role of interleukin-6 in monitoring severe case of coronavirus disease 2019. EMBO Mol Med [Internet]. 2020 Jul 7 [cited 2023 Feb 26];12(7). Available from: https://pubmed.ncbi.nlm.nih.gov/32428990/

67.Sabaka P, Koščálová A, Straka I, Hodosy J, Lipták R, Kmotorková B, *et al.* Role of interleukin 6 as a predictive factor for a severe course of Covid-19: retrospective data analysis of patients from a long-term care facility during Covid-19 outbreak. BMC Infect Dis [Internet]. 2021 Dec 1 [cited 2023 Feb 26];21(1):1–8. Available from: https://bmcinfectdis.biomedcentral.com/articles/10.1186/s12879-021-05945-8

68.Chen LYC, Hoiland RL, Stukas S, Wellington CL, Sekhon MS. Assessing the importance of interleukin-6 in COVID-19. Lancet Respir Med [Internet]. 2021 Feb 1 [cited 2023 Feb 26];9(2):e13. Available from: http://www.thelancet.com/article/S2213260020306007/fulltext

69.Potere N, Batticciotto A, Vecchié A, Porreca E, Cappelli A, Abbate A, *et al.* The role of IL-6 and IL-6 blockade in COVID-19. Expert Rev Clin Immunol [Internet]. 2021 [cited 2023 Feb 26];17(6):601–18. Available from: https://pubmed.ncbi.nlm.nih.gov/33874829/

70.Grifoni E, Valoriani A, Cei F, Lamanna R, Gelli AMG, Ciambotti B, *et al.* Interleukin-6 as prognosticator in patients with COVID-19. Journal of Infection. 2020 Sep;81(3):452–82.

71.Zhang ZL, Hou YL, Li DT, Li FZ. Laboratory findings of COVID-19: a systematic review and meta-analysis. Scand J Clin Lab Invest. 2020 Oct 1;80(6):441–7.

72.McGonagle D, Sharif K, O'Regan A, Bridgewood C. The Role of Cytokines including Interleukin-6 in COVID-19 induced Pneumonia and Macrophage Activation Syndrome-Like Disease. Autoimmun Rev [Internet]. 2020 Jun 1 [cited 2023 Feb 26];19(6). Available from: https://pubmed.ncbi.nlm.nih.gov/32251717/

73.Fu B, Xu X, Wei H. Why tocilizumab could be an effective treatment for severe COVID-19? J Transl Med [Internet]. 2020 Apr 14 [cited 2023 Feb 26];18(1). Available from: https://pubmed.ncbi.nlm.nih.gov/32290839/

74.Zhang C, Wu Z, Li JW, Zhao H, Wang GQ. Cytokine release syndrome in severe COVID-19: interleukin-6 receptor antagonist tocilizumab may be the key to reduce mortality. Int J Antimicrob Agents [Internet]. 2020 May 1 [cited 2023 Feb 26];55(5). Available from: https://pubmed.ncbi.nlm.nih.gov/32234467/

75.Lipworth B, Chan R, Lipworth S, RuiWen Kuo C. Weathering the Cytokine Storm in Susceptible Patients with Severe SARS-CoV-2 Infection. J Allergy Clin Immunol Pract [Internet]. 2020 Jun 1 [cited 2023 Feb 26];8(6):1798–801. Available from: https://pubmed.ncbi.nlm.nih.gov/32311489/

76.Mangalmurti N, Hunter CA. Cytokine Storms: Understanding COVID-19. Immunity [Internet]. 2020 Jul 14 [cited 2023 Feb 26];53(1):19–25. Available from: https://pubmed.ncbi.nlm.nih.gov/32610079/

77.Haywire Immune Response Eyed in Coronavirus Deaths, Treatment - WSJ [Internet]. [cited 2023 Feb 26]. Available from: https://www.wsj.com/articles/haywire-immune-response-eyed-in-coronavirus-deaths-treatment-11586430001

78.Schulert GS, Grom AA. Pathogenesis of macrophage activation syndrome and potential for cytokine- directed therapies. Annu Rev Med [Internet]. 2015 Jan 14 [cited 2023 Feb 26];66:145–59. Available from: https://pubmed.ncbi.nlm.nih.gov/25386930/

79.Aziz M, Fatima R, Assaly R. Elevated interleukin-6 and severe COVID-19: A meta-analysis. J Med Virol. 2020 Nov 2;92(11):2283–5.

80.Herold T, Jurinovic V, Arnreich C, Lipworth BJ, Hellmuth JC, von Bergwelt-Baildon M, *et al.* Elevated levels of IL-6 and CRP predict the need for mechanical ventilation in COVID-19. Journal of Allergy and Clinical Immunology. 2020 Jul;146(1):128-136.e4.

81.Patil S, Gondhali G, Acharya A. "Serial ferritin titer" monitoring in COVID-19 pneumonia: valuable inflammatory marker in assessment of severity and predicting early lung fibrosis — prospective, multicentric, observational, and interventional study in tertiary care setting in India. Egypt J Intern Med. 2022 Dec 12;34(1):75.

82.Dahan S, Segal G, Katz I, Hellou T, Tietel M, Bryk G, *et al.* Ferritin as a Marker of Severity in COVID-19 Patients: A Fatal Correlation. Isr Med Assoc J [Internet]. 2020 Aug 1 [cited 2023 Feb 25];22(8):494–500. Available from: https://europepmc.org/article/MED/33236582

83.Chen R, Sang L, Jiang M, Yang Z, Jia N, Fu W, *et al.* Longitudinal hematologic and immunologic variations associated with the progression of COVID-19 patients in China. Journal of Allergy and Clinical Immunology. 2020 Jul;146(1):89–100.

84.Zhou F, Yu T, Du R, Fan G, Liu Y, Liu Z, *et al.* Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. The Lancet. 2020 Mar;395(10229):1054–62.

85.Ren B, Yan F, Deng Z, Zhang S, Xiao L, Wu M, *et al.* Extremely High Incidence of Lower Extremity Deep Venous Thrombosis in 48 Patients With Severe COVID-19 in Wuhan. Circulation [Internet]. 2020 Jul 14 [cited 2023 Feb 25];142(2):181–3. Available from: https://pubmed.ncbi.nlm.nih.gov/32412320/

86.Liu Y, Du X, Chen J, Jin Y, Peng L, Wang HHX, *et al.* Neutrophil-to-lymphocyte ratio as an independent risk factor for mortality in hospitalized patients with COVID-19. Journal of Infection. 2020 Jul;81(1):e6–12.

87.Qin C, Zhou L, Hu Z, Zhang S, Yang S, Tao Y, *et al.* Dysregulation of Immune Response in Patients With Coronavirus 2019 (COVID-19) in Wuhan, China. Clinical Infectious Diseases. 2020 Jul 28;71(15):762–8.

88. Lippi G, Cervellin G. Procalcitonin for diagnosing and monitoring bacterial infections: for or against? Clinical Chemistry and Laboratory Medicine (CCLM). 2018 Jul 26;56(8):1193–5.

89. Lippi G, Plebani M. Procalcitonin in patients with severe coronavirus disease 2019 (COVID-19): A meta-analysis. Clinica Chimica Acta. 2020 Jun;505:190–1.

90. Hu R, Han C, Pei S, Yin M, Chen X. Procalcitonin levels in COVID-19 patients. Int J Antimicrob Agents. 2020 Aug;56(2):106051.

91. Cuesta M, Thompson CJ. The syndrome of inappropriate antidiuresis (SIAD). Best Pract Res Clin Endocrinol Metab. 2016 Mar;30(2):175–87.

92. Ellison DH, Berl T. The Syndrome of Inappropriate Antidiuresis. New England Journal of Medicine. 2007 May 17;356(20):2064–72.

93. Ho KS, Narasimhan B, Kumar A, Flynn E, Salonia J, El-Hachem K, *et al.* Syndrome of inappropriate antidiuretic hormone as the initial presentation of COVID-19: A novel case report. Nefrología. 2021 Mar;41(2):219–20.

94. Yousaf Z, Al-Shokri SD, Al-soub H, Mohamed MFH. COVID-19-associated SIADH: a clue in the times of pandemic! American Journal of Physiology-Endocrinology and Metabolism. 2020 Jun 1;318(6):E882–5.