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Original Research Article

A study on diagnostic and prognostic significance of laboratory parameters and its correlation with HRCT of lung in Covid-19 patients

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ABSTRACT

Introduction: Coronaviruses had a wide range of symptoms from asymptomatic to severe illness and has caused significant morbidity and death among large population infected with this virus.

Aims and Objectives: To correlate CT severity score with each blood parameter like (TLC counts, Absolute neutrophil count, Absolute lymphocyte count, neutrophilic/lymphocytic ratio, quantitative D-dimer, ferritin, CRP, IL-6 and LDH). To assess diagnostic and prognostic significance of each parameter in predicting severity of disease.

Materials and Methods: The present study was a 1-year prospective, a hospital-based study from 1 August 2020 to 31 July 2021 and included 100, COVID-19 confirmed cases. The clinical, radiological and laboratory reports were collected. The CT score were compared with different blood parameters. The results were analyzed statistically using correlation coefficient(r).

Results and Observations : 27 patients having a CT severity score 1-10 were graded in mild category. 27 patients having CT severity score 11-15 were graded in moderate category. 34 patients having CT severity score 16- 20 were graded as severe. 12 patients having CT severity score 21-25 were graded as very severe. The present study showed raised TLC count > 11000 / μ l in 35% patients suffering from COVID -19. Absolute neutrophil count > 7000/ μ l was seen in 50 % patients. 65% patients showed Absolute lymphocyte count <1000> 3.5 was seen in 83% patients. Raised D-Dimer >500 ng/ml was observed in 74 % patients. 92 % subjects showed raised Serum ferritin >150ng/ml. IL-6 >10pg/ml was seen in 92 % patients. Q-CRP >10 mg/l was seen in 97 % patients.

Conclusion: HRCT chest severity score of patients with COVID-19 has positive correlation with various laboratory parameters and hence can be used as an indicator of disease severity. It can play a vital role in the management of COVID-19 and should be used for comprehensive evaluation.

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1. Introduction

The coronavirus disease 2019 (Covid 19) is also known as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). It had a wide range of symptoms from asymptomatic to severe illness and has caused significant morbidity and death among large population infected with

this virus. Therefore, various studies have been done to identify the diseased patient early to prevent poor clinical outcome. The present study is different from the other studies since it evaluates all the biochemical and haematological parameters which play a significant role in the prognosis of the patient and the findings compared with the HRCT score. The other researches conducted deal with a few prognostic markers only.

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2. Materials and Methods

The current study is done on patients who were admitted to the COVID care center, Govt Medical College Kota, Rajasthan, between 1st August 2020 to 31 July 2021 and tested positive for COVID-19 on RT-PCR reaction assay using the viral nucleic acid detection kit recommended by the National Institute of Virology. Nasopharyngeal and oropharyngeal swabs were collected from suspected patients into collection tubes containing virus transport medium for extraction of total viral RNA.

The clinical history, signs and symptoms, HRCT scan, and laboratory reports of the patients were recorded. Inflammatory biomarkers utilized in this study were Total leucocyte count, Absolute neutrophil count, Absolute lymphocyte count, Neutrophil-Lymphocyte ratio, IL-6, Q-CRP, serum ferritin, Lactate Dehydrogenase and D-dimer.

The covid 19 cases were categorized into mild, moderate, and severe, depending upon HRCT score. The signs of ground-glass opacity, consolidation, fibrosis, and air trapping were analysed quantitatively using a radiologic scoring system ranging from 0-25 points. Each lobe of the lung was evaluated by 0-5 points, on the basis of the area involved. The scoring was done as 0 for normal performance, 1 for less than 5% area involved, 2 for 6%-25%, 3 for 26%-50%, 4 for 51%-75%, and 5 for more than 75%. A total score was eventually recorded by the addition of the scores for each lobe.¹ An association and correlation was made between HRCT scoring with each parameter of baseline investigations to study the severity of the disease.

2.1. Statistical analysis

Statistical analysis was done by using Pearson's correlation coefficient (r) between the CT severity score and each quantitative variable of investigations.

Approval from the ethical committee was obtained, and informed consent was taken from all patients.

3. Results

A total of 100 patients, confirmed as COVID-19 positive by RT-PCR test, and had undergone HRCT scan procedure and blood investigations (CBC, S. ferritin, IL-6, Q-CRP, LDH, D-dimer) were included. Males (59%) were more commonly affected than females (41%). The most common clinical symptoms was shortness of breath (61%) followed by fever (51%), and cough (38%).

Distribution of COVID-19 patients according to CT severity score, and percentage of subjects with various parameters were studied. Patients having raised TLC count with mild CT scores were 13 out of 27 (48.14%), and moderate CT scores were 8 out of 27(29.62%), in severe CT scores they were 12 out of 34 (35.29%), in very severe CT score were 2 out of 12(35%). HRCT Score and TLC count showed a positive correlation (r=0.07).

Table 1: Shows the normal range of different laboratory parameters.

Parameter	Normal level
TLC	4000-11000/ μ l
ANC	2000-7000/ μ l
ALC	1000-3000/ μ l
S. Ferritin	15-150 mcg/l
Neutrophil/Lymphocyte	0.78-3.53
D-Dimer	<500ng/ml
IL-6	0-16.4 Pg/ml
LDH	240-480 IU/l
CRP	<10 mg/l

Table 2: Showing the distribution of study subjects according to CT severity score

Grade	Ct score	Number of patients
Mild	1-10	27
Moderate	11-15	27
Severe	16-20	34
Very severe	21-25	12

Table 3: Distribution of COVID -19 patients with abnormal laboratory parameters

Laboratory parameters	Number of patients with increased/decreased
TLC>11000/ μ l	35/100
ANC>7000/ μ l	50/100
ALC<1000/ μ l	65/100
N/L Ratio>3.5	83/100
D-Dimer>500 ng/ml	74/100
Serum ferritin> 150ng/ml	92/100
IL-6>10pg/ml	92/100
Q-CRP>10 mg/l	97/100
LDH>400 IU/L	91/100

Raised ANC counts in patients with mild CT scores were 14 out of 27 (51.85%), in moderate CT scores were 12 out of 27(44.44%), in severe CT scores were 21 out of 34 (61.76%) and in very severe CT score (21-25) were 3 out of 12(25%). HRCT Score and ANC count showed a positive correlation (r= 0.01)

Patients with decreased ALC count in mild CT scores were 13 out of 27 (48.14%), in moderate CT scores were 17 out of 27(62.69%), in severe CT scores were 25 out of 34 (73.52%), in very severe CT score were 10 out of 12(83.33%). HRCT Score and ALC count shows a negative correlation (r -0.25).

Patients with raised N/L ratio in mild CT score were 22 out of 27 (81.48%), in moderate CT scores were 20 out of 27(70.07%), in severe CT score were 34 out of 34 (100%), in very severe CT scores were 7 out of 12(58.33%). HRCT Score and N/L ratio showed positive correlation (r= 0.08).

Patients with raised D-Dimer in mild CT score were 15 out of 27 (55.55%), in moderate CT scores were 24 out of

27(88.88%), in severe CT score were 28 out of 34 (82.35%), in very severe CT scores were 7 out of 12(58.33%). HRCT Score and D-Dimer showed a positive correlation ($r=0.11$).

Patients with raised S. Ferritin in mild CT scores were 20 out of 27 (74.07%), in moderate CT scores were 27 out of 27(100%), in severe CT score were 33 out of 34 (97.05%), in very severe CT scores were 12 out of 12(100%). HRCT Score and S. Ferritin showed positive correlation ($r=0.83$).

Patients with raised IL-6 in mild CT score were 20 out of 27 (74.07%), in moderate CT scores were 26 out of 27(96.29%), in severe CT score were 34 out of 34 (100%), in very severe CT scores were 12 out of 12(100%). HRCT Score and IL-6 showed positive correlation ($r=0.44$).

Patients with raised Q-CRP in mild CT score were 24 out of 27 (88.88%), in moderate CT scores were 27 out of 27(96.29%), in severe CT score were 34 out of 34 (100%), in very severe CT scores were 12 out of 12(100%). HRCT Score and Q-CRP showed positive correlation ($r=0.27$).

Patients with raised LDH in mild CT score were 21 out of 27 (77.77%), in moderate CT score (11-15) were 27 out of 27(100%), in severe CT score (16-20) were 34 out of 34 (100%), in very severe CT scores were 9 out of 12(75%). HRCT Score and LDH showed positive correlation ($r=0.26$).

4. Discussion

Covid 19 is a highly infectious and severe inflammatory disease caused by SARS-CoV-2. Mostly this infection causes leukocytopenia and increase in lymphocyte count. But the immunological phenotype of severe COVID-19 infection is like bacterial infection characterized by elevated total leukocyte count mainly neutrophilic and decreased lymphocytes. The neutrophils to lymphocytes ratio correlate well with severity of the disease.

In COVID-19 infection, the mechanism of increased neutrophil count is not clearly described although the two most common mechanisms of the increased neutrophil count are as follows.

1. Neutrophils express toll-like receptors TLR-7/8/9 which are involved in the detection of SARS-CoV2
2. SARS -CoV enters the cells of the nasopharynx by ACE-2 receptors which bind to porphyrin and causes accumulation of Fe²⁺. This led to high ferritin levels which further causes high serum LDH, low lymphocyte count and platelets, and increased WBC and neutrophils.²

In the present study, TLC was increased in 35% of cases and showed a positive correlation ($r=0.07$) with CT severity score. Similar studies done by Sharma S. et al.³ and Agarwal N et al.⁴ showed increased TLC count in 48.7% and 39.03% respectively, concluding a rising trend of leukocyte counts with increasing disease severity.

Absolute neutrophil count was increased in 50% of cases and also showed a positive correlation ($r=0.01$) with CT severity score. These findings match with similar studies done by Agarwal N et al.⁴ (62.62%), Chandran J et al.⁵

Absolute lymphocyte count was found to be decreased in 65% of cases and showed a negative correlation ($r=-0.25$) with CT severity score. Similar studies have been done by various other authors, Sharma S. et al.,³ Ahmad Z et al.⁶, Xie Y et al.⁷ Agarwal N et al.,⁴ Chandran J et al.⁵ indicating that when the absolute lymphocyte count decreases, the patient's condition also deteriorates.

In the present study, Neutrophil/Lymphocyte ratio was increased in 83% of cases and showed a positive correlation($r=0.08$) with CT severity score. Chandran J et al.⁵ Man MA et al.,⁸ Kurri N at al.,⁹ Citu C et al.,¹⁰ showed that there was a significant positive correlation between NLR and CT severity score (i.e., NLR increases with the severity of lung involvement).

We also studied various inflammatory markers which play a significant role in predicting the prognosis of patients with covid 19 infection. Serum Ferritin, LDH, IL-6, and CRP were evaluated. All were found to be increased and had a positive correlation with the CT severity score. In covid 19 infection the macrophages are activated and release IL-6, IL-10, and ferritin. This leads to hyper-ferritinemia, which in turn, produces a vicious cycle by secreting several pro-inflammatory (IL-1 β) and anti-inflammatory cytokines (IL-10) as well as macrophage activation.¹⁰

The present study also highlighted similar findings as S.ferritin was raised in 92% of cases and showed a positive correlation ($r=0.83$) with CT severity score. Various authors also studied these and reported similar findings. They are Padiya G S et al.,¹¹ Kömürçüoğlu B et al.,¹² Sharma S. et al.,³ Vidyashree S et al.,¹³ Gupta P et al.,¹⁴ Padiya G S et al.¹¹ did a study on 59 patients which included 21 patients in the mild group,31 in the moderate group, and 7 in the severe group. CT severity score was found to be positively correlated with ferritin levels ($p < 0.001$). Strong positive correlations were found between CT scores and serum ferritin in the mild ($r = 0.84$), Moderate ($r = 0.92$), and severe groups ($r = 0.082$). Gupta P et al.¹⁴ suggested that a combination of ferritin and isolated levels of inflammatory markers can help in assessing the severity of Covid – 19 diseases.

Lactate dehydrogenase (LDH) is found in almost all the tissues of the body including those in the blood, heart, kidney, brain, and lungs. It is found in high concentrations in lung tissue (isozyme 3) and therefore patients with severe COVID-19 pneumonia have high LDH levels in the circulation often progressing to acute respiratory distress syndrome (ARDS). LDH isoenzymes catalyze the interconversion of pyruvate and lactate, which is a crucial step in the anaerobic metabolism of glucose when oxygen is unavailable or limited. In SARS-CoV2 due to decreased O₂

supply to body tissue, LDH is released from various body tissues due to increase proliferation and death of cells.¹⁴ Wu MY et al.¹⁵ reported that LDH levels in severe COVID-19 patients were higher than those in non-severe patients. He also reported that an increase or decrease of LDH value indicates the progress or improvement of the patient.

Serum LDH was raised in 91% of cases in the current study and demonstrated a strong positive correlation ($r=0.26$) with the CT severity score. Similar studies were done by Xiong Y et al.,¹⁶ Kömürçüoğlu B et al.¹² They also agreed that LDH has a significantly positive correlation with the severity of pneumonia assessed on initial CT. Tordjman M et al.¹⁷ reported that with a cut-off value of LDH > 380, severe pneumonia on CT can be detected with 100% sensitivity, and in our study also 100% sensitivity was detected with a cut-off value of 400.

CRP is an acute phase reactant produced by the liver during an inflammatory response and enhances the phagocytosis of microorganisms through specific CRP receptors. In COVID-19 pneumonia various cytokines are released and stimulate hepatocytes to produce CRP.

In the present study, Quantitative CRP was raised in 97% of cases ($r=0.27$). It showed marked increase in severe CT score cases and had 100% sensitivity in moderate and severe cases. In mild cases, CRP of three patients was within the normal range. This may be due to differences in the timing of investigations. Sharma S. et al.³ Lonsane A et al.¹⁸ Francone M et al.¹⁹ showed statistically significant correlation between CT score vs CRP ($p<0.0001$, $r=0.6204$). Tan C. et al.²⁰ reported that CRP in severe patients increased significantly at the initial stage of the disease and CRP predicted early Covid-19 disease. Huang Y. et al.²¹ concluded that CRP was significantly elevated in a non-survival group compared to the survival group of Covid-19 patients.

Interleukin-6 is a pleiotropic cytokine produced in response to tissue damage by viral infections. The cytokine storm caused by its release damages the lung and leads to ARDS in many severe cases. This IL-6 level might be an important tool to detect the severity of COVID-19 infection and further decide treatment protocol.²²

IL-6 was raised in 92% of our cases indicating a good association between IL6 and CT score. It showed a positive correlation ($r=0.44$) with the CT severity score. However, in 08 cases IL6 was within normal ranges which belonged to the mild and moderate groups. This can be due to a delay in the inflammatory response in these patients or the IL6 test being done before the CT scan. Similar studies done by Bhandari S et al.²³ showed high IL-6 levels, exhibited same severity of illness ($P < 0.001$) and marked lung involvement ($P < 0.05$) in covid 19 patients. Ahmed A et al.²⁴ observed that serum Interlukin-6 levels were significantly raised in 97 patients with mean levels of 20.43 ± 19.66 (pg./ml) thus concluding Interlukin-6 as an important biomarker for

diagnosis and predicting the severity of COVID-19. Chen et al.²⁵ reported similar findings with significant elevation of inflammatory cytokine IL-6 levels in critically ill COVID-19 patients

The D-dimer level is used in patients to detect thrombosis. Studies have reported an increase in D-dimer and fibrinogen concentrations in the early stages of COVID-19 disease. A 3 to 4-fold rise in D-dimer levels is linked to poor prognosis. This occurs due to activation and damage to endothelial cells by covid 19 virus which expresses the ACE-2 receptor, thereby increasing prothrombotic factors.

In the present study, D-dimer was increased in 74% of cases and showed a positive correlation ($r=0.11$) but sensitivity was less compared to other biomarkers. Francone M et al.¹⁹ did a similar study and reported that statistically significant correlation was found between CT score vs D-dimer ($p<0.0001$, $r=0.6625$) levels. Shifa Y et al.²⁶ found increased D-dimer levels (>500 ng/mL) in 92.3% of patients. Zhou and his colleague.²⁶ conducted a retrospective study involving 191 COVID-19 patients and found that elevated D-dimer at admission was a risk factor for the death of adult patients. However, this conclusion was not consistent in other studies.

5. Conclusion

Our study has concluded that, the HRCT chest severity score of patients with COVID-19 is positively correlated with laboratory parameters like Total leucocyte count, Absolute neutrophil count, Neutrophil/ Lymphocyte ratio, D-dimer, Serum ferritin, IL-6, Q-CRP, Serum LDH and hence can be used as an indicator of disease severity. Absolute lymphocyte count showed a negative correlation with CT severity score. HRCT chest imaging with baseline investigations (TLC, ANC, N/L, ALC, S. D-dimer, S. Ferritin, IL-6, Q-CRP and S.LDH) played a vital role in the management plan for COVID-19 and should be used for comprehensive evaluation. These tests should be used in conjunction with the results of nucleic acid tests, HRCT findings, clinical findings, and other vital laboratory parameters. The findings in the present study are comparable with those reported in the literature. However, a few of the data taken from the literature varies from our studies.

6. Limitations

The present study has been conducted on a limited number of patients for it to be of valuable significance. A properly timed sequential study and follow-up carried out on higher number of patients would be truly informative and of greater and valid significance. Some blood investigations in our study were done before HRCT, some were done after HRCT, also some investigations were done inside the hospital and some were done outside the hospital, breaking

uniformity in test results, all these tests should ideally be carried out in a single place. Due to the constantly changing nature of each strain of virus, there is a change in clinical presentation and the severity of involvement of each organ. Therefore, tests carried out may not be very specific for each strain of COVID-19. With every change of variant, parameters decided by tests carried out during their morbidity will prove to be of ultimate significance.

7. Conflict of Interest

None.

8. Source of Funding

None.


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