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An association between immune status and chest CT scores in COVID-19 patients

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Abstract

Background: The characteristic features of the immune responses of COVID-19 patients and how they reflect lung involvement have not been clearly elucidated.

Aim: The aim of this study was to examine the immune status and the correlations thereof with chest CT scores and lung involvement of patients with COVID-19.

Methods: In this retrospective and single-center study, 72 patients with laboratory-confirmed COVID-19 were recruited. The counts of peripheral lymphocyte subsets ($CD3^+$ T cells, $CD4^+$ T cells, $CD8^+$ T cells, $CD19^+$ B cells and $CD16^+$ 56^+ NK cells) and those of serum immunoglobulins (IgA, IgG, IgM) were measured and their associations with chest CT scores analysed.

Results: The proportions of lymphopenia in patients with extensive lung involvement were twice that in the general study population. In the severe disease group, the levels of total lymphocytes, T cells, B cells, NK cells; and serum IgA levels, were significantly lower than in the mild disease group (all $P < .05$). We found that the numbers of lymphocyte subsets and the IgA level negatively correlated with the chest CT scores. On multivariate regression analysis, pretreatment decreases in total lymphocytes, $CD3^+$ T cells, $CD4^+$ T cells, and $CD19^+$ B cells, and serum IgA levels, were independent predictors of severe lung involvement.

Conclusions: The cell numbers of peripheral lymphocyte subsets and the serum IgA level were negatively correlated with the chest CT scores in COVID-19 patients. These parameters tended to independently predict severe lung involvement in such patients.

1 | INTRODUCTION

The coronavirus disease 2019 (COVID-19) pandemic, which first emerged in December 2019 in pneumonia patients in Wuhan, China, and then spread worldwide, has caused over 150 million confirmed cases and more than 3 million deaths to date.[1,2] The clinical course of COVID-19 disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) may vary from asymptomatic to severe; the latter patients may require mechanical ventilation.[3] The incidence of severe COVID-19 disease is between 15.7% to 26.1%. The criteria include clinical tachypnoea and hypoxia, as well as an extent of lung infiltration >50%.[4] Reverse transcription-polymerase chain reaction (RT-PCR), which is the main test used to diagnose COVID-19, has certain limitations in terms of sample collection and transport, and kit performance. The sensitivity is relatively low (60%-71%). Chest CT is routinely used to image and diagnose all types of pneumonia, affording high sensitivity (97%) when (rapidly) diagnosing COVID-19 disease.[5] Severe COVID-19 patients have more extensive lung involvement than asymptomatic or mild patients. In addition, there are significant positive correlations between COVID 19 severity evident on chest CT and dyspnoea and deteriorating gas exchange.[4,6]

Approximately 20% of patients with COVID-19 develop acute respiratory distress syndrome (ARDS), the clinical course of which exhibits uncontrolled immune system activation, excessive cytokine release and multi-organ failure.[7,8] Increased serum levels of proinflammatory cytokines are associated with pulmonary inflammation and extensive lung injury.[9] Lymphocyte subsets ($CD4^+$ T cells, $CD8^+$ T cells, B cells and natural killer [NK] cells) play very important roles in the immune response. Changes in lymphocyte subsets vary by the type of viral infection and viral pathogenic mechanisms.[10] However, the characteristic features of the immune response in patients with severe COVID-19 infections and their effects on the clinical findings remain unclear. Some studies have suggested that immune system overactivation plays a role in the immunopathology[11]; others have stated that loss or dysfunction of immune cells may be responsible.[12]

In this study, we examined the relationship between the severity of lung involvement and immune system factors (which vary widely among COVID-19 patients). We aimed to characterise immune status, which can illuminate the pathogenesis of COVID-19, and sought an early indicator of severe lung involvement in patients with COVID-19 infections.

What's known

- The severity of lung involvement varies considerably among patients in the acute phase of COVID-19.
- Severe COVID-19 patients have more extensive lung involvement than asymptomatic or mild patients.
- The difference in the extent of lung involvement among COVID-19 patients and its relation to the patient's immune status during acute infection is poorly understood.

What's new

- This is a pioneering study demonstrating a significant relationship between severe lung involvement and host immune status during the acute phase of COVID-19.
- The cell numbers of peripheral lymphocyte subsets and the serum IgA levels were negatively correlated with the chest CT scores in COVID-19 patients.
- These parameters tended to independently predict severe lung involvement in such patients.
- Early evaluation of CT scores and immune parameters in COVID-19 patients is essential in determining severe patients.
- Thus, the patient groups that need to be applied aggressive treatment modalities in the early stages may be determined, and a decrease in the morbidity and mortality rates because of COVID-19 may be achieved.

2 | MATERIAL AND METHODS

2.1 | Study population

This retrospective cohort study was conducted in Necmettin Erbakan University Meram Medical Faculty Hospital, which is a large tertiary hospital. The study was approved by the local Ethics Committee of Necmettin Erbakan University Meram Faculty of Medicine (decision no. 2020/2620); the study adhered to all relevant tenets of the Declaration of Helsinki (1975). Among patients who were hospitalised with a diagnosis of COVID-19 (laboratory-confirmed cases, detected by the SARS-CoV-2 RNA molecular method) between March 10, 2020, and June 08, 2020, those who met the following inclusion criteria were accepted into the study: (a) Over 18 years of age; (b) Had undergone chest CT without contrast; and, (c) Laboratory analyses of immunoglobulin levels and the numbers of cells in various peripheral lymphocyte subgroups had been performed within 72 hours after admission. The exclusion criteria were as follows: (a) Severe motion artifacts on CT; (b) The use of drugs (corticosteroids, anticonvulsants, cyclosporine and other immunosuppressive treatments) or the presence of a disease (an immune deficiency, celiac disease, inflammatory bowel disease, a malignancy or an autoimmune disease) at the time of COVID-19 diagnosis that might affect the levels of serum immunoglobulins and peripheral lymphocytes; and, (c) A diagnosis of a structural lung disease (bronchiectasis, an interstitial lung disease, etc). A total of 117 patients were enrolled according to the inclusion criteria but 45 were excluded for the reasons as shown in Figure 1. As a result, 72 confirmed COVID-19 patients were finally enrolled in the study.

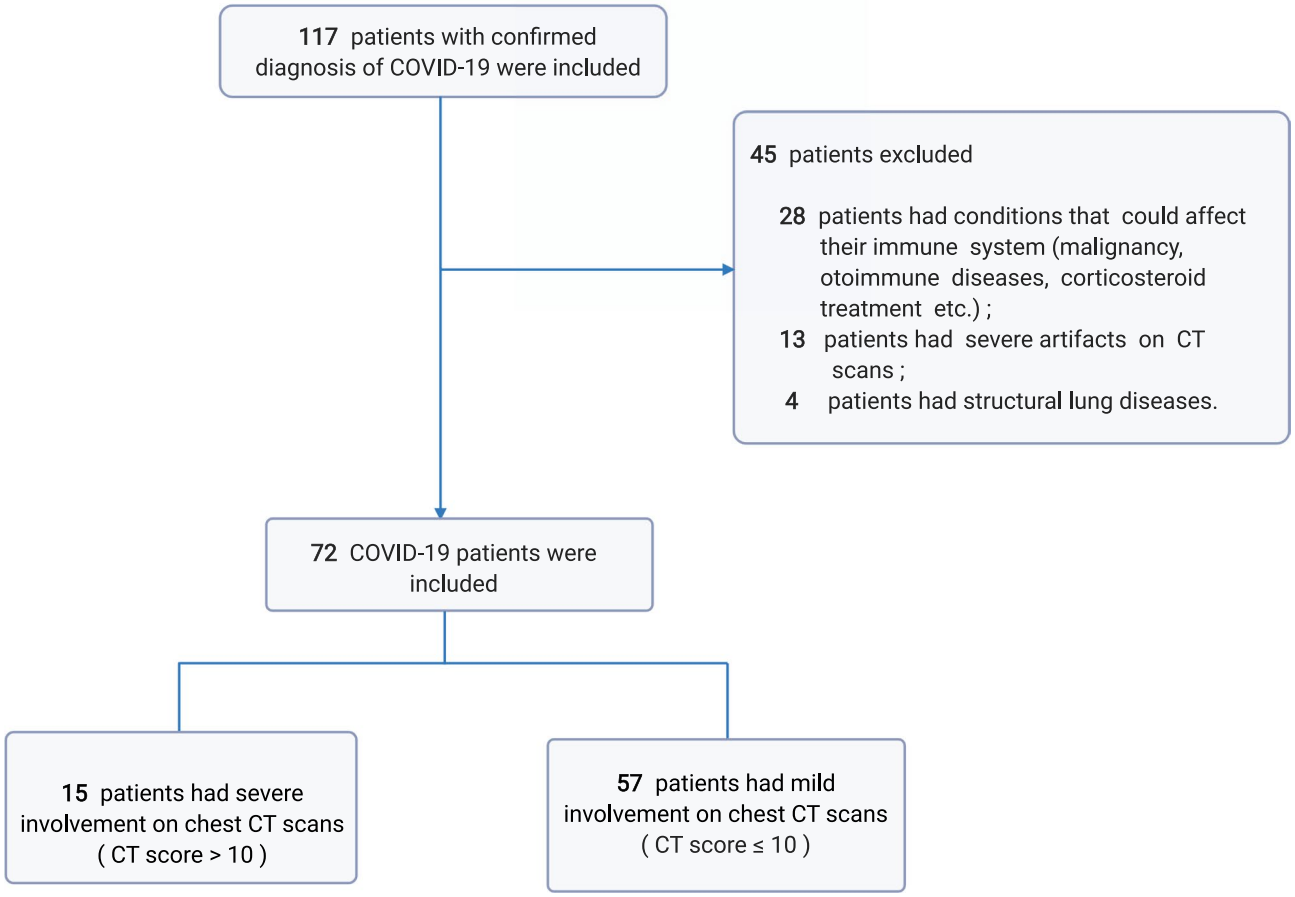

FIGURE 1 Flow chart of patient screening. The inclusion and exclusion criteria were strictly applied throughout screening

2.2 | Data collection

We recorded age, gender, medical history, laboratory values, chest CT findings and disease severity assessments at the time of admission to our hospital, from electronic medical records. Patients with COVID-19 are considered to have severe illness if they exhibit an oxygen saturation (SpO2) <94% on room air at sea level, a respiratory rate >30 breaths/min, a ratio of the arterial partial pressure of oxygen to the fraction of inspired oxygen (PaO_2/FiO_2) <300 mmHg, or lung infiltration >50%.[13] We investigated the relationship between the severity of lung involvement evident on chest CT and the numbers of total lymphocytes and those of various subgroups of peripheral lymphocytes (T-cells, B-cells, NK cells), and serum immunoglobulin levels (IgG, IgA and IgM) in COVID-19 patients.

2.3 | SARS-CoV-2 RNA detection

SARS-CoV-2 RNA was detected using the reverse transcription-polymerase chain reaction (RT-PCR) test; we combined nasopharyngeal and oropharyngeal swabs. RNA was isolated using the vNAT solution (Bioeksen, Istanbul, Turkey). A Rotor-Gene Q (Qiagen, Antwerp, Belgium) instrument and a Biospeedy SARS-CoV-2 RT-qPCR kit (Bioeksen) were used in all tests. The data were analysed using Rotor-Gene Q Software.

2.4 | Serum immunoglobulin measurements

Serum immunoglobulin levels were determined by nephelometric methods (Siemens BNII System, Erlangen, Germany).

2.5 | Flow cytometric analysis

Samples of EDTA-anticoagulated peripheral blood (2 mL) were collected from all patients before initial treatment. All samples were tested within 6 hours of being obtained. $CD3^+/CD4^+/CD8^+$ T cell, $CD19^+$ B cell and $CD16^+$ $CD56^+$ NK-cell counts (cells/μL) were measured via multiple-color flow cytometry using human monoclonal anti-CD3-fluorescein isothiocyanate (FITC)-, anti-CD4-phycoerythrin (PE)-, anti-CD8-allophycocyanin (APC)-, anti-CD19-PE-, anti-CD16-APC- and anti-CD56-PE-conjugated antibodies (the BD Multitest) according to the manufacturer's instructions. The cells were analysed on a BD FACS Canto II flow cytometry system (BD Biosciences, Erembodegem, Belgium). Absolute lymphocyte counts were determined as the products of the white blood cell (WBC) count and the

percentage lymphocyte proportion as measured by the Mindray BC-6200 autohematology analyzer (Nanshan, Shenzhen, China).

2.6 | Chest CT and CT image review

All chest CT examinations were performed using a Somatom Drive (Siemens Healthineers) scanner. Unenhanced CT scans were obtained from the bilateral apex to the base for all patients. The scanning parameters were as follows: Tube voltage, 120 kV; tube current-time product, 50-100 mAs; pitch, 0.6; matrix, 512 × 512; slice thickness, 3 mm, and reconstructed slice thickness, 1.5 mm. Images were viewed on a Syngo Via Workstation (Siemens Healthineers) within the lung window setting (width: 1,000-1,500 Hounsfield units [HU], level: 700-550 HU) and mediastinal window setting (width: 300-350 HU, level: 30-40 HU). All CT images were evaluated by two radiologists (working in consensus) with 12 and 25 years of thoracic imaging experience.

Severity scores were awarded to each of the five lung lobes: 0 for no involvement (0%), 1 for minimal involvement (1%-25%), 2 for mild involvement (26%-50%), 3 for moderate involvement (51%-75%) and 4 for advanced involvement (76%-100%). The total severity score was the sum of the scores of the five lobes. We categorised all patients into two groups according to their total chest CT scores (≤10 vs >10): Mild involvement and severe involvement (Figures 2 and 3).

2.7 | Statistical analysis

Continuous variables are presented as medians with interquartile ranges (IQRs), and categorical variables as numbers and percentages in each category. We used the Mann-Whitney U-test to evaluate continuous data and the chi-squared test to evaluate categorical data when comparing the mild and severe COVID-19 patients. Spearman's correlation test was used to assess the association between lymphocyte subset counts, serum immunoglobulin levels, and the chest CT scores. Univariate and multivariate logistic regression analyses were used to identify risk factors for severe lung involvement. All variables with *P*-values <.1 on univariate analysis were entered into forward, stepwise multivariate logistic regression analysis. The SPSS statistical package (ver. 22.0; IBM Corp., Armonk, NY, USA) and GraphPad Prism statistical software (ver. 8.0; GraphPad Software, San Diego, CA, USA) were used for all analyses. A *P*-value <.05 was considered statistically significant.

3 | RESULTS

3.1 | General characteristics of COVID-19 patients

We included 72 patients with COVID-19. The median age was 48 years (IQR, 38-65 years) and 33 (46%) were female. We found no between-gender difference in disease prognosis or the extent of pulmonary involvement (*P* = .396 and 0.280 respectively). Nineteen patients had severe disease, of whom 15 exhibited extensive lung involvement on chest CT (>50% lung involvement, CT score >10). The lymphopenia proportion was 32% (n = 23) in a general study population, 63% (n = 12) in patients with severe disease and 67% (n = 10) in those with extensive lung involvement. As shown in Table 1, in patients with severe COVID-19, the total CT score, the creatinine, D-dimer and ferritin levels, the erythrocyte sedimentation rate (ESR), and the C-reactive protein (CRP) level were higher than in patients with mild COVID-19 (*P* = <0.001, 0.001, 0.003, 0.017, 0.039 and 0.001 respectively).

3.2 | Immunological indicators and COVID-19 severity

We categorised 19 patients (26%) as severely ill on admission. These patients had significantly lower levels of total lymphocytes (*P* < .001), $CD3^+$ T cells (*P* < .001), $CD3^+$ $CD4^+$ T cells (*P* < .001), $CD3^+$ $CD8^+$ T cells (*P* < .001), $CD19^+$ B cells (*P* = .013) and $CD16^+CD56^+$ NK cells (*P* = .024) than patients with mild COVID-19. The serum IgA levels were significantly lower in patients with severe compared with mild illness (*P* = .028). We found no between-group IgG or IgM difference (*P* = .157 and 0.922 respectively; Table 1).

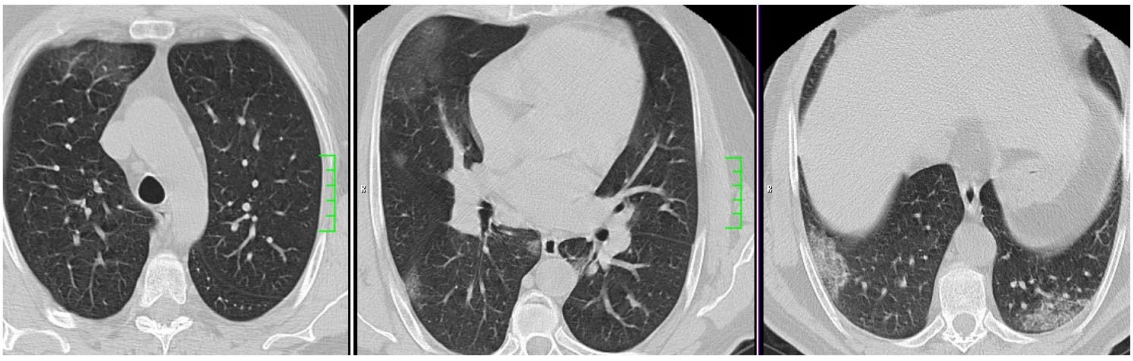

FIGURE 2 52-year-old man with COVID-19 pneumonia. Axial CT images shows bilateral, multilobar, peripheral and central, patchy ground-glass opacities. COVID-19 chest CT severity score is "7"

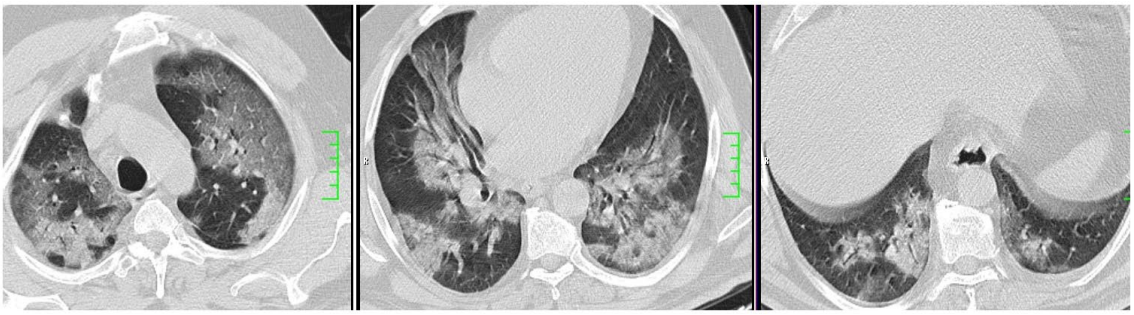

FIGURE 3 56-year-old man with COVID-19 pneumonia. Axial CT images shows bilateral, multilobar, peripheral and central, patchy ground-glass opacities. COVID-19 chest CT severity score is "15"

TABLE 1 Demographic and laboratory results of mild and severe COVID-19 patients

	COVID-19 prognosis			
	Normal range	Mild (n = 53)	Severe (n = 19)	*P*
Age (y)	—	46 (35-61)	50 (42-67)	.216*
Gender				
Female (n = 33)		25 (47%)	8 (42%)	.396**
Male (n = 39)		28 (53%)	11 (58%)	.396
Total CT score	0	3 (1-4)	7 (3-11)	<.001
WBC count (cell/μL)	4000-10 000	5910 (4625-8065)	6390 (5020-10 065)	.661
NEU count (cell/μL)	1500-7300	3860 (2660-5585)	4385 (2950-8620)	.219
PLT count (cell/μL)	150 000-400 000	218 500 (156 500-280 500)	196 000 (152 000-251 000)	.326
Total LYM count (cell/μL)	1000-4800	1645 (1188-1978)	880 (665-1258)	<.001
$CD3^+$ count (cell/μL)	723-2737	1326 (864-1554)	579 (413-857)	<.001
$CD3^+$ $CD4^+$ count (cell/μL)	404-1612	710 (417-932)	331 (204-524)	<.001
$CD3^+$ $CD8^+$ count (cell/μL)	220-1129	407 (299-627)	264 (189-320)	<.001
$CD19^+$ count (cell/μL)	80-616	136 (88-266)	59 (41-107)	.013
$CD16^+$ $CD56^+$ count (cell/μL)	84-724	151 (101-232)	99 (63-146)	.024
IgG (mg/dL)	700-1600	1145 (980-1300)	996 (877-1248)	.157
IgA (mg/dL)	70-400	202 (151-259)	131 (81-238)	.028
IgM (mg/dL)	40-230	91 (70-172)	106 (56-162)	.922
AST (U/L)	15-40	23 (18-41)	26 (21-37)	.238
ALT (U/L)	10-45	25 (18-35)	28 (20-38)	.309
Creatinine (mg/dL)	0.7-1.2	0.85 (0.70-1.10)	1.30 (0.90-1.55)	.001
D-dimer (ng/mL)	0-243	114 (75-500)	641 (198-818)	.003
Ferritin (ng/mL)	30-250	128 (39-500)	470 (81-864)	.017
ESR (mm/1 h)	0-20	21 (14-47)	44 (18-62)	.039
CRP (mg/L)	0-10	14 (5-36)	52 (16-94)	.001

Abbreviations: CT, computed tomography; WBC, white blood cells; NEU, neutrophils; PLT, platelets; LYM, lymphocytes; AST, aspartate aminotransferase; ALT, alanine aminotransferase; ESR, erythrocyte sedimentation rate; CRP, C-reactive protein.

*Mann-Whitney *U* test (data were shown as medians with interquartile ranges [IQRs]).; **Chi Square test (data were shown as number and percentages).

3.3 | Immunological parameters and chest CT scores in patients with COVID-19

We found severe lung involvement on chest CT (>50% lung involvement, CT score >10) in 15 patients (21%). Compared with patients with mild lung involvement, the severe cases exhibited significantly lower levels of total lymphocytes ($P < .001$), $CD3^+$ T cells ($P < .001$), $CD3^+$ $CD4^+$ T cells ($P = .001$), $CD3^+$ $CD8^+$ T cells ($P = .007$), and $CD19^+$ B cells ($P = .003$) and IgA (<0.001). The levels of $CD16^+CD56^+$ NK cells, and IgG and IgM, did not differ

between the groups (P = .358, 0.150 and 0.096 respectively) (Figure 4).

The numbers of total lymphocytes (r = −.4241, P < .001), $CD3^+$ T cells (r = −.4248, P < .001), $CD3^+CD4^+$ T cells (r = −.4338, P < .001), $CD3^+CD8^+$ T cells (r = −.3013, P = .01), and $CD19^+$ B cells (r = −.3689, P = .001); and the IgA level (r = −.2891, P = .036), were negatively correlated with the chest CT scores. The numbers of $CD16^+C56^+$ NK cells, and the IgG and IgM levels, exhibited no such correlations (P = .12, .12 and .509 respectively; Figure 5).

3.4 | Immunological factors associated with severe lung involvement in COVID-19 patients

Table 2 shows the results of univariate and multivariate logistic regression analyses. We found that lower counts of total lymphocytes, $CD3^+$ T cells, $CD3^+$ $CD4^+$ T cells and $CD19^+$ B cells; and a lower IgA level, were risk factors for severe lung involvement on univariate regression analysis. All variables with P-values <.1 on univariate analysis were entered into forward, stepwise multivariate logistic regression analysis, which showed that lower counts of total lymphocytes (odds ratio [OR] = 3.296, 95% confidence interval [CI] 1.308-11.674, P = .016), $CD3^+$ T cells (OR = 5.758, 95% CI 1.823-17.382, P = .003), $CD3^+CD4^+$ T cells (OR = 4.171, 95% CI 1.452-14.605, P = .011), and $CD19^+$ B cells (OR = 2.539, 95% CI 1.145-7.316, P = .028) and a lower IgA level (OR = 2.347, 95% CI 1.173-13.891, P = .031), were risk factors for severe lung involvement in COVID-19 patients.

4 | DISCUSSION

In the current study, the proportions of lymphopenia in patients with severe disease and extensive lung involvement were twice that in the general study population. In the severe disease group, the numbers of total lymphocytes, T cells, B cells and NK cells, and the serum IgA levels, were significantly lower than in the mild disease group.

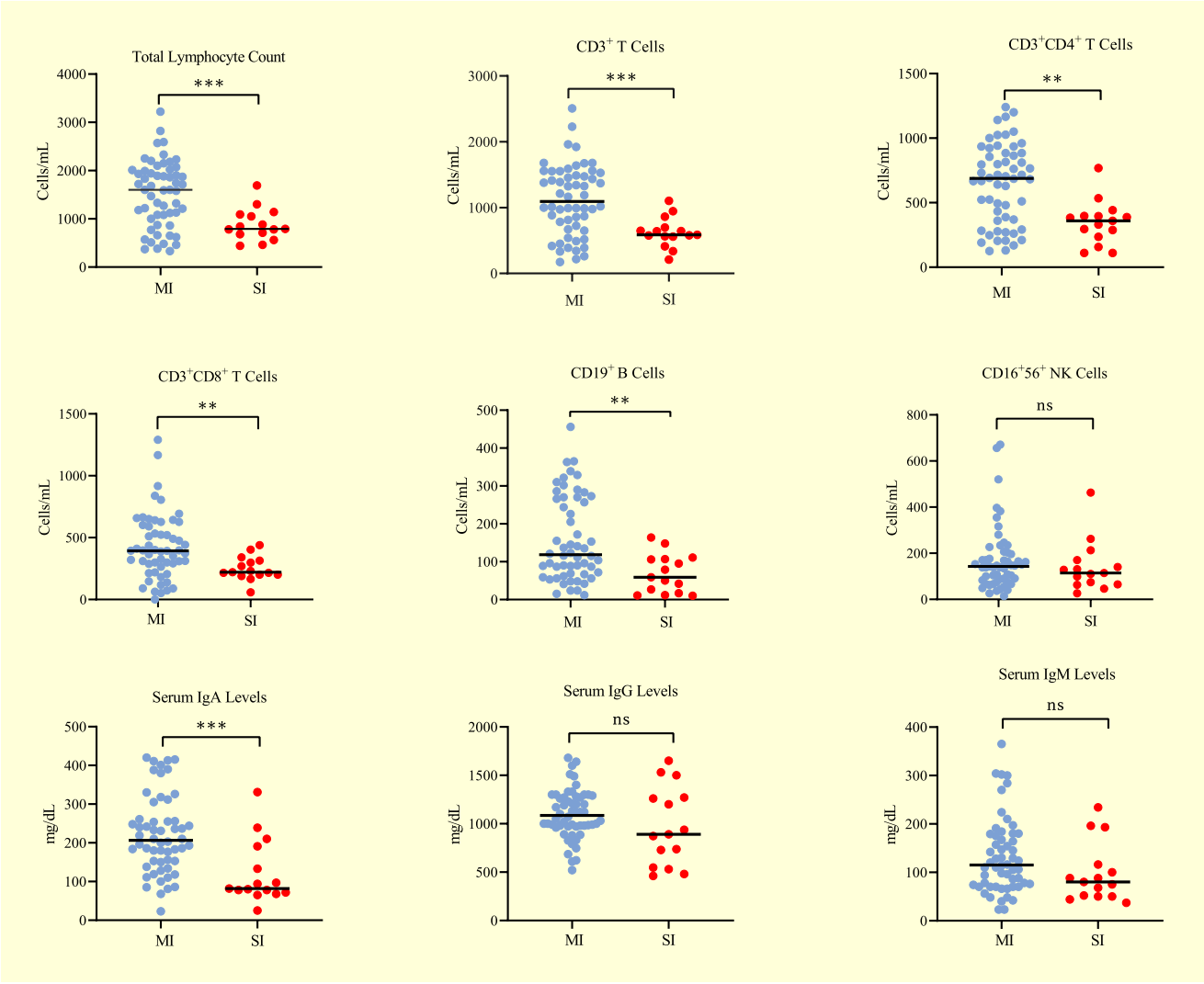

FIGURE 4 Comparison of peripheral lymphocyte subset counts and serum immunoglobulin levels between patients with COVID-19 exhibiting mild (lung) involvement (MI) and severe involvement (SI) on chest CT. ***P < .001; **P < .01; ns, not significant

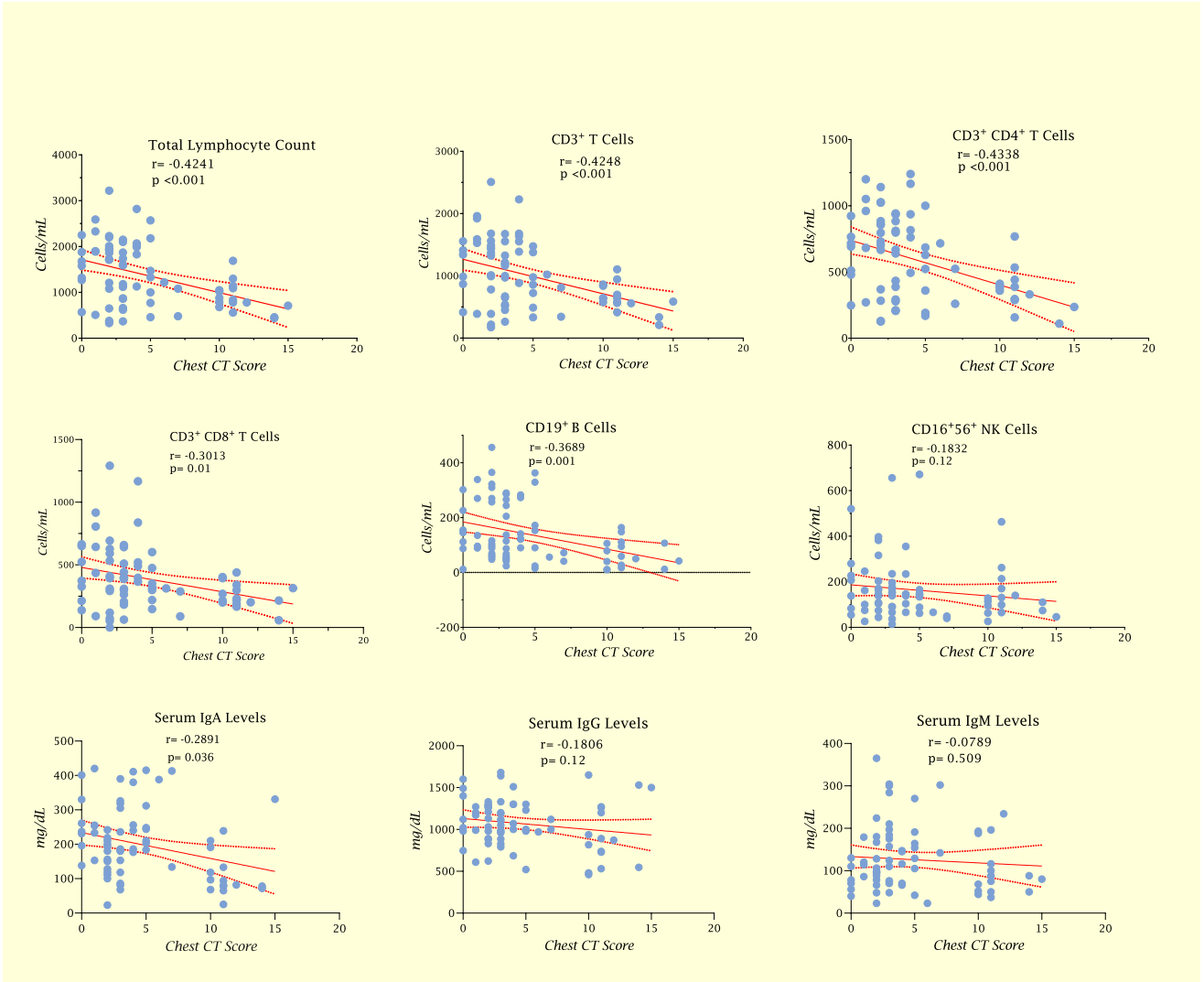

FIGURE 5 Correlations between the numbers of various types of peripheral lymphocytes, serum immunoglobulin levels and chest CT scores. Solid line: fitted curve; dashed line: 95% confidence interval (CI) of the fitted curve

TABLE 2 Logistic regression analysis results of risk factors for severe lung involvement in COVID-19 patients

Variables	OR	95% CI	*P*	OR	95% CI	*P*
Total Lym count (<1000 vs ≥1000 cell/μL)	5.385	1.641-17.664	**.005**	3.296	1.308-11.674	**.016**
$CD3^+$ count (<723 vs ≥723 cell/μL)	8.786	2.433-31.725	**<.001**	5.758	1.823-17.382	**.003**
$CD3^+$ $CD4^+$ count (<404 vs ≥404 cell/μL)	7.312	2.049-26.102	**.002**	4.171	1.452-14.605	**.011**
$CD3^+$ $CD8^+$ count (<220 vs ≥220 cell/μL)	2.513	0.782-8.074	.122			
$CD19^+$ count (<80 vs ≥80 cell/μL)	4.062	1.264-13.056	**.010**	2.539	1.145-7.316	**.028**
$CD16^+$ CD 56^+ count (<84 vs ≥84 cell/μL)	1.331	0.393-4.504	.646			
IgG (<700 vs ≥700 mg/dL)	1.232	0.497-11.058	.137			
IgA (<70 vs ≥70 mg/dL)	6.692	1.242-21.480	**.023**	2.347	1.173-13.891	**.031**
IgM (<40 vs ≥40 mg/dL)	0.849	0.082-8.767	.849			

Abbreviations: OR, odds ratio; CI, confidence interval; Lym, lymphocyte.

Bold indicates *P*-values <.05 are statistically significant.

We found that decreases in the numbers of total lymphocytes, the vast majority of lymphocyte subsets (except for NK cells), and lower IgA levels were associated with high COVID-19 chest CT scores and extensive lung involvement.

Lymphocytes are vital in terms of maintaining immune system functionality. As observed in patients with immune system disorders, viral and other infections can trigger pathological variations in lymphocyte levels.[14] Similar to the findings of our study, it has been reported in previous studies that dramatic decreases in total lymphocyte counts were common in COVID-19 patients and were associated with the severity of chest CT findings and poor disease prognosis. It has been shown that COVID-19 patients with extensive lung involvement exhibit more severe decreases in lymphocyte levels, and a negative correlation was evident between lymphocyte counts and lesions in the lungs.[15,16] In our prenent study, 63% of severe COVID-19 patients and 67% of those with extensive lung involvement had lymphopenia. In previous studies, the incidence of lymphopenia in patients with COVID-19 was 72% [10], 84% in SARS-CoV patients[17] and 34% in MERS-CoV patients.[18] Some authors have suggested causes of the development of lymphopenia in COVID-19 patients. The first suggestion is that lymphopenia may develop as a result of mobilisation of peripheral circulating lymphocytes to the inflamed areas of damaged lung tissues.[19] The dramatic decrease in lymphocyte numbers observed in COVID-19 patients may be related to direct invasion by SARS-CoV-2. The virus may deplete lymphocytes, triggering disorders of the immune response. This mechanism is in play in MERS-CoV infection. Also, autoimmune antibodies triggered by viral agents may trigger hematopoietic apoptosis and deficiencies in the production and differentiation of various types of lymphocytes.[15,20] Increased cytokine levels and lymphopenia in COVID-19 patients are indicators of poor prognosis. The high cytokine levels in these patients may cause the development of lymphopenia. IL-1, IFN-γ and IL-6 inhibit T lymphocyte proliferation. Proinflammatory cytokines inhibit lymphopoiesis while inducing granulopoiesis in the bone marrow of patients with SARS-CoV-2 infection. As a result, the increased numbers of monocytes and granulocytes produce more inflammatory cytokines, creating a vicious circle that deepens the lymphopenia and worsens the clinical condition.[21]

CD3, a biological marker of mature T lymphocytes, mediates activation of $CD4^+$ T cells and $CD8^+$ T cells.[22] $CD4^+$ and $CD8^+$ T cells play important roles in the stability and co-ordination of the immune system.[23] When SARS-CoV-2 first enters tissues such as respiratory epithelial cells, viral peptides are presented to $CD8^+$ cytotoxic T cells via the class I major histocompatibility complex (MHC). The $CD8^+$ T cells become activated and engage in clonal expansion. They then mediate the development of virus-specific effector and memory cells. $CD8^+$ T cells lyse virus-infected cells using cytotoxic molecules such as perforin and granzyme B. Shortly after this acute response, viral particles are recognised by professional antigen presenters (dendritic cells and macrophages) and presented to $CD4^+$ cells via the MHC class II.[24]

Differentiation of naive $CD4^+$ T cells into effector and memory cells is one of the main functions of T cell-mediated immunity. An appropriate balance between naive and memory $CD4^+$ T cells is essential for effective maintenance of the immune response. This balance is disrupted in critically ill COVID-19 patients in favor of naive $CD4^+$ T cells. It has also been reported that the viral load of SARS-CoV-2 is negatively correlated with the numbers of $CD4^+$ and $CD8^+$ T cells.[25] Given the decrease in the number of T lymphocytes in COVID-19 patients, SARS-CoV-2 can directly invade T lymphocytes and accelerate apoptosis of such cells by activating the intrinsic and extrinsic apoptosis pathways.[20] Fas (CD95) is the cell surface receptor of the tumor necrosis factor superfamily, and is also a death receptor that mediates T lymphocyte apoptosis. It has been reported that CD95 expression by $CD4^+$ and $CD8^+$ T cells is increased in COVID-19 patients compared with healthy controls, and correlated negatively with the numbers of such cells.[26]

NK cells are innate lymphoid cells that become activated without any need for antigen stimulation, kill infected cells directly, and contribute to the activation and co-ordination of the adaptive immune system.[27] Recent studies reported low NK cell levels in severe COVID-19 patients compared with mild cases.[28-30] Besides, in this patient group, the number of NK cells may fall, and their functions may become impaired. It has been shown that COVID-19 patients have higher levels of the CD94/NK group 2 member A (NKG2A) receptor, which produces inhibitory signals and suppresses cytokine secretion and the cytotoxic actions of NK cells, compared with healthy individuals. In such patients, high NKG2A levels are accompanied by low levels of the activation markers CD107a, IFNɣ, IL-2, and TNFα of T and NK cells.[28] As was true of NKG2A, it was shown that the programmed cell death protein 1 (PD-1) was expressed at a higher level in T cells of patients with COVID-19 than healthy controls. It has been suggested that the levels of PD-1, T-cell immunoglobulin and mucin domain-3 (Tim-3) increase as COVID-19 symptoms become exacerbated and T cells are depleted.[31]

The B cell response to viral infection protects against acute infection and provides immunity to recurrent infections. After the infection ceases, antibodies with serological memory continue to be secreted by plasma cells. Memory B cells formed during the initial infection can rapidly respond to recurrent infections by forming high-affinity plasma cells. Long-term protection is achieved via the stimulation of long-lived plasma cells and memory B cells.[32] The peripheral blood of COVID-19 patients contains fewer naive B cells and more plasma cells than does the blood of healthy controls.[33] B cell immunity is critical if cytopathic viruses are to be cleared. SARS-CoV-2 elicits a fast and robust, specific B cell response. Virus-specific IgM, IgG and IgA antibodies are detected in the circulation within a short time after infection.[34]

Antibodies play essential roles in the immune responses to viruses; patients with antibody-deficient diseases such as common variable immunodeficiency (CVID) are predisposed to viral infections.[35] IgA is the most abundant antibody in the body, being a critical component of the mucosal immunity defense against infectious agents that target the mucosa.[36] Ig A binds to both bacteria and viruses, preventing their adhesion to epithelial cells, thus contributing to the first-step immune response against various pathogens.[37] IgA

is principally responsible for the mucosal immunity of the respiratory and gastrointestinal systems, which are the main routes of entry of SARS-CoV-2 and other pathogens. IgA can neutralise the virus by preventing SARS-CoV-2 from reaching and adhering to epithelial cells.[38] In COVID-19 patients with abnormally low IgA levels, the risk of pneumonia was 4-fold higher than that of other patients.[39] In one study investigating the relationship between the prevalence of selective immunoglobulin A deficiency (SIgAD) and the prevalence of COVID-19 in various countries worldwide, it was concluded that a strong positive correlation was evident. In Japan, where SIgAD is extremely rare, the frequency of infections by, and mortality from, SARS-CoV-2 is low. In the USA and other Western countries, where the frequency of SIgAD is much higher than in Japan, the infection and death rates from SARS-CoV-2 are higher than in Japan.[37]

Our study had some limitations. First, the immune parameters of patients with COVID-19 were examined only in the acute period of the disease. It would have been better to examine peripheral lymphocyte subgroups and immunoglobulins in the acute phase and after recovery from disease. The second limitation is that that memory T and B cells were not examined. Measuring the levels of memory cells specific for the disease is important when it is sought to understand the immunopathogenesis of COVID-19. In addition, it would be meaningful to determine the levels of the various lymphocyte subgroups, not only their activities and functions. Finally, the viral load of SARS-CoV-2 was not measured using molecular methods, and any correlation thereof with the levels of immune parameters was not examined. Such correlations would be useful, especially given the widespread lung involvement evident on CT, and the (common) severe pneumonia.

Despite these limitations, we showed that the severity of lung involvement, which may vary considerably among patients in the acute phase of COVID-19, and thus the chest CT scores, were related to immune parameter measurements. To the best of our knowledge, very few studies on this subject have appeared. For patients with COVID-19, no prior study has examined the relationship between IgA levels and chest CT scores. In this respect, our study offers a new perspective; it may be possible to develop nasal vaccines that increase IgA levels in mucosal membranes, neutralising SARS-CoV-2 in mucosae before the virus enters the cell.

5 | CONCLUSIONS

The prevalence and prognosis of SARS-CoV-2 pneumonia vary considerably among individuals. We have shown that the prognosis and the severity of chest CT involvement in COVID-19 are related to the levels of immune cells. Interestingly, we found that the levels of B lymphocytes, IgA levels and T lymphocyte levels were negatively correlated with the chest CT scores. Also, the levels of total lymphocytes, $CD3^+$ T cells, $CD3^+$ $CD4^+$ T cells, and $CD19^+$ B cells and serum IgA levels, were lower than the normal ranges, and associated with severe lung involvement. Early evaluation of CT scores and immune parameters in COVID-19 patients is essential in determining severe patients. Thus, the patient groups that need to be applied aggressive treatment modalities in the early stages may be determined, and a decrease in the morbidity and mortality rates because of COVID-19 may be achieved. New treatment strategies that improve immune system cell numbers and functions should be considered, especially for severely COVID-19 patients.

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DISCLOSURES

All authors declare that they do not have a conflict of interest.

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