Clinical Characteristics and Predictive Value of low CD4+T Count in Patients with Moderate and Severe COVID-19: A Multicenter Retrospective Study

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Research article

Keywords: CD4+T cells, COVID-19, In-hospital death, SARS-CoV-2

DOI: https://doi.org/10.21203/rs.3.rs-46009/v1

Abstract

Background

In December 2019, coronavirus disease 2019 (COVID-19) caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) emerged in Wuhan, Hubei, China. And, it has become a global pandemic. Describe the patient's clinical symptoms in detail, finding markers that predict the prognosis of patients with COVID-19 are of great value.

Methods

In this multicenter, retrospective study, 476 patients with COVID-19 were recruited from a consecutive series. After screening, a total of 395 patients were included in this study. All-cause death was the primary endpoint. All patients were followed up from admission till discharge or death.

Results

The dominant symptoms observed in the study included fever on admission, cough, fatigue and shortness of breath. The most frequent comorbidities were hypertension and diabetes. Compared with patients with higher $CD4^+$T cells level, patients with lower $CD4^+$T cells level were older and were more frequently male. In terms of laboratory findings, lymphocyte count, $CD4^+$T cell count, $CD8^+$T cell count were significantly lower in low group than in higher group. The case in-hospital death rate was significant higher in patients with lower $CD4^+$T level. After adjusting for potential confounding factors, $CD4^+$T count below the lower limit of normal showed independent prognostic value for all-cause in-hospital death in patients with COVID-19.

Conclusions: Reductions in lymphocytes and lymphocyte subsets are common in COVID-19 patients, especially in severe cases. It is the $CD8^+$T count, not the $CD4^+$T count, that reflected the severity of the patient's disease. Lower $CD4^+$T count is independently associated with an increased rate of in-hospital death.

Trial registration: Prognostic Factors of Patients With COVID-19, NCT04292964. Registered 03 March 2020. https://clinicaltrials.gov/ct2/show/NCT04292964.

1. Background

In December 2019, an outbreak of coronavirus disease 2019 (COVID-19), an acute respiratory illness caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was detected in mainland China. Although the overall case fatality rate of patients with COVID-19 is relatively low [1], the number of deaths related to COVID-19 has already exceeded the sum of SARS and MERS, which has brought great harm to human beings. Moreover, the fatality rate of patients with severe COVID-19 is higher and the

harm is bound to be greater [2]. Describe the patient's clinical symptoms in detail, finding markers that predict the prognosis of patients with COVID-19 are of great value.

The decline of T-lymphocytes in peripheral blood is a typical laboratory characteristic of patients with COVID-19, especially in severe patients [3, 4]. A recent study recruited 21 patients with COVID-19 including 11 severe patients and 10 moderate patients. The study showed absolute number of T-lymphocytes, $CD4^+T$ and $CD8^+T$ cells decreased in almost all the patients, and significantly lower in severe patients (294.0, 177.5 and 89.0 × 10^6/L) than moderate patients (640.5, 381.5 and 254.0 × 10^6/L). Meanwhile, most patients did not show a decrease in B-lymphocytes count, but showed a tendency to an increased B-lymphocytes count. This phenomenon suggested that SARS-CoV-2 infection may primarily affect T-lymphocytes particularly $CD4^+T$ and $CD8^+T$ cells [4]. T-lymphocytes play a critical role in antiviral immunity. $CD4^+T$ lymphocyte subsets secretes high level of effector cytokines, especially interferon-γ (IFN-γ), which are essential for virus clearance [5, 6]. Previous study also showed that the drastic reduction in total lymphocytes indicated the consumed immune cells and the destructed cellular immune function by coronavirus [7]. However, there are not enough studies on whether $CD4^+T$ predicts the prognosis of COVID-19 patients.

2. Methods

2.1 Subjects

Medical records from 476 patients with confirmed COVID-19 were collected in Hubei General Hospital and Chongqing Three Gorges Central Hospital. Missing $CD4^+T$ or CD^+8T data (n = 58), malignant tumor (n = 8), younger than 18 years (n = 11), eGFR ≤ 30 ml/min (n = 3), and pregnant (n = 1) were excluded. Finally, 395 patients were analyzed in this study (Fig. 1). The positive infected cases were confirmed by testing new coronavirus nucleic acid by real-time fluorescent Polymerase Chain Reaction (RT-PCR). Patients with severe COVID-19 were defined according to the New Coronavirus Pneumonia Prevention and Control Program issued by the National health commission of the People's Republic of China (5th edition). Patients with respiratory distress (respiratory rates ≥ 30 per/min or resting oxygen saturation ≤ 93% or partial pressure of arterial oxygen (PaO2)/inspired oxygen fraction (FiO2) ≤ 300 mmHg or respiratory failure requiring mechanical ventilation, were defined as severe COVID-19, and the remaining patients were defined as moderate patients. $CD4^+T$ count, $CD8^+T$ count and lymphocyte count were divided into lower group and higher group according to the low value of laboratory reference values. The study was a multicenter, retrospective, observational registry with clinicaltrials.gov identifier NCT04292964. All study procedures were approved by the local ethics committee (approval NO. 20200701). All data were collected by experienced researchers using blinded methods.

2.2 Baseline Data And Follow-up

Demographic and clinical characteristics were collected from the electronic medical record system. Data collection of laboratory results were defined by the results of the first test after admission. All patients in the study were followed up from admission till death or discharge. The outcome was defined as the in-hospital death rate.

2.3 Statistical Analysis

Continuous data were expressed as mean ± standard deviation (SD) or median (interquartile range) according to the distribution. Categorical variables were presented as frequency rates with percentages. Continuous variables with normal distribution were compared using independent group T-test; otherwise, the Mann-Whitney U test. Categorical data were tested using the Chi-square test and Fisher's exact Chi-square test. Cox proportional-hazards models were used to perform univariate analyses and multivariate analyses to identify the association between $CD4^+$T count and in-hospital death. Kaplan-Meier survival analysis with log-rank test was performed to estimate the cumulative survival rate of groups with higher or lower $CD4^+$T count. Statistical analyses were performed by the IBM SPSS Statistics 26.0 software. P (two-sided) value less than 0.05 was considered statistical significance.

3. Results

3.1 Baseline characteristics

Baseline characteristics are shown in Table 1. The average age was 53 years, and 53 (51.6%) were male. Among these cases, fever on admission (263, 66.6%) was the most common symptom. Cough, shortness of breath, fatigue, and sputum production were present in 257 patients (65.1%), 118 patients (29.9%), 107 patients (27.2%), and 102 patients (25.9%), respectively. Headache (36, 9.1%), nausea or vomiting (36, 9.1%), myalgia or arthralgia (34, 8.6%), sore throat (22, 5.6%), and chill (7, 6.7%) were rare in our study. The most frequent comorbidities were hypertension (102, 25.8%) and diabetes (47, 11.3%). The proportion of coronary heart disease, hepatitis B infection, and chronic obstructive pulmonary disease was 6.4% (25/392), 2.3% (9/392), and 1.5% (6/392), respectively.

Table 1

Baseline characteristics of different degrees of $CD4^+$ T cell in all patients.

Variables	All(N = 395)	CD4 + T: lower than the normal low limit (N = 195)	CD4 + T: higher than the normal low limit (N = 200)	*P*	Missing date
Baseline					
Male/female(n)	204/191	115/80	89/111	0.004	
Age(years)	53.1 ± 15.7	55.0 ± 16.5	51.3 ± 14.8	0.033	
Temperature (℃)	36.8 (36.5–37.3)	36.9 (36.6–37.6)	36.8 (36.5–37.1)	0.036	31 (7.8%)
Heart rate (min)	85.0 (77.0–94.0)	85.0 (78.0–96.0)	84.5 (76.0–92.0)	0.103	4 (1.0%)
SBP (mmHg)	126.0 (116.0-136.0)	126.0 (115.0-136.5)	126.0 (117.0-136.0)	0.577	6 (1.5%)
DBP (mmHg)	78.0 (70.0–85.0)	76.0 (70.0–85.0)	78.0 (71.0–85.0)	0.741	6 (1.5%)
Symptoms and signs—No, %					
Fever on admission	263 (66.6%)	141 (72.3%)	122 (61.0%)	0.017	
Nasal congestion	2 (0.5%)	2 (1.0%)	0 (0%)	0.243	
Headache	36 (9.1%)	20 (10.3%)	16 (8.0%)	0.436	
Cough	257 (65.1%)	138 (70.8%)	119 (59.5%)	0.019	
Sore throat	22 (5.6%)	10 (5.1%)	12 (6.0%)	0.706	
Sputum production	102 (25.9%)	56 (28.9%)	46 (23.0%)	0.184	1 (0.3%)
Fatigue	107 (27.2%)	59 (30.4%)	48 (24.0%)	0.153	1 (0.3%)

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; COPD, Chronic obstructive pulmonary disease; WBC, white blood cell count; Hb, Hemoglobin; PLT, platelet count; LYM, lymphocyte; ALT, alanine aminotransferase; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin.

Variables	All(N = 395)	CD4 + T: lower than the normal low limit (N = 195)	CD4 + T: higher than the normal low limit (N = 200)	*P*	Missing date
Shortness of breath	118 (29.9%)	75 (38.5%)	43 (21.5%)	< 0.001	
Nausea or vomiting	36 (9.1%)	23 (11.8%)	13 (6.5%)	0.068	
Myalgia or arthralgia	34 (8.6%)	20 (10.3%)	14 (7.0%)	0.249	
Chill	12 (3.0%)	8 (4.1%)	4 (2.0%)	0.223	
Throat congestion	3 (0.8%)	0 (0%)	3 (1.5%)	0.248	
Coexisting disorders—No, %					
Diabetes	47 (11.9%)	22 (11.3%)	25 (12.5%)	0.709	
Hypertension	102 (25.8%)	48 (24.6%)	54 (27.0%)	0.588	
Coronary heart disease	25 (6.4%)	15 (7.7%)	10 (5.1%)	0.277	3 (0.8%)
Hepatitis B infection	9 (2.3%)	6 (3.1%)	3 (1.5%)	0.334	3 (0.8%)
COPD	6 (1.5%)	5 (2.6%)	1 (0.5%)	0.119	3 (0.8%)
Laboratory findings					
WBC ($\times 10^9$/L)	5.3 (4.2-7.0)	5.0 (3.8-7.0)	5.6 (4.5-7.0)	0.008	2 (0.5%)
Hb (g/L)	131.0 (118.5–143.0)	132.0 (117.0-143.0)	129.0 (120.0-142.3)	0.809	2 (0.5%)
PLT ($\times 10^9$/L)	189.0 (145.5–252.0)	160.0 (129.0-214.0)	220.5 (170.0-364.0)	< 0.001	2 (0.5%)
LYM ($\times 10^9$/L)	1.1 (0.8–1.5)	0.8 (0.6-1.0)	1.5 (1.2–1.8)	< 0.001	6 (1.5%)

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; COPD, Chronic obstructive pulmonary disease; WBC, white blood cell count; Hb, Hemoglobin; PLT, platelet count; LYM, lymphocyte; ALT, alanine aminotransferase; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin.

Variables	All(N = 395)	CD4 + T: lower than the normal low limit (N = 195)	CD4 + T: higher than the normal low limit (N = 200)	*P*	Missing date
LYM < 1.1 × 10^9/L	199 (51.2%)	163 (84.5%)	27 (13.8%)	< 0.001	6 (1.5%)
ALT (U/L)	23.0 (15.0–39.0)	24.1 (15.4–38.8)	22.0 (15.0–39.0)	0.388	4 (1.0%)
Cr (umol/L)	64.0 (53.0–78.0)	66.5 (56.0–79.0)	61.0 (50.0–77.0)	0.005	5 (1.3%)
D-dimer (mg/L)	0.43 (0.24–0.99)	0.50 (0.28–1.12)	0.38 (0.22–0.84)	0.023	14 (3.5%)
K (mmol/L)	4.0 (3.7–4.3)	4.0 (3.6–4.3)	4.1 (3.7–4.3)	0.243	6 (1.5%)
Hs-CRP (mg/L)	5.0 (2.2–22.9)	8.2 (5.0-48.5)	4.9 (1.1-7.0)	< 0.001	45 (11.4%)
PCT (ng/ml)	0.05 (0.03–0.08)	0.06 (0.04–0.11)	0.04 (0.02–0.06)	< 0.001	21 (5.3%)
$CD4^+$T cells count	410.0 (265.0-567.0)	262.0 (188.0-325.0)	564.0 (478.5–716.0)	< 0.001	
$CD8^+$T cells count	246.0 (154.0-348.0)	168.0 (107.0-250.0)	322.0 (244.3-443.5)	< 0.001	
CD4/CD8 ratio	1.6 (1.2–2.2)	1.4 (1.1–1.9)	1.8 (1.4–2.3)	< 0.001	
Abnormalities on chest CT—No,%					
Ground-glass opacity	170 (48.7%)	78 (46.7%)	92 (50.5%)	0.473	46 (11.6%)
Local patchy shadowing	135 (38.7%)	71 (42.5%)	64 (35.2%)	0.159	46 (11.6%)
Treatment					
Oxygen inhalation	328 (84.3%)	174 (90.2%)	154 (78.6%)	0.002	6 (1.5%)

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; COPD, Chronic obstructive pulmonary disease; WBC, white blood cell count; Hb, Hemoglobin; PLT, platelet count; LYM, lymphocyte; ALT, alanine aminotransferase; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin.

Variables	All(N = 395)	CD4 + T: lower than the normal low limit (N = 195)	CD4 + T: higher than the normal low limit (N = 200)	*P*	Missing date
Glucocorticoids	94 (23.8%)	64 (32.8%)	30 (15.0%)	< 0.001	
Antiviral treatment	388 (98.2%)	191 (97.9%)	197 (98.5%)	0.721	
Intravenous immunoglobulin	71 (18.2%)	37 (19.2%)	34 (17.3%)	0.625	5 (1.3%)
Antibiotic treatment	179 (45.3%)	112 (57.4%)	67 (33.5%)	< 0.001	
Antifungal treatment	4 (1.0%)	2 (1.0%)	2 (1.0%)	1.000	
	Clinical outcome				
Death (No,%)	27 (6.8%)	25 (12.8%)	2 (1.0%)	< 0.001	

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; COPD, Chronic obstructive pulmonary disease; WBC, white blood cell count; Hb, Hemoglobin; PLT, platelet count; LYM, lymphocyte; ALT, alanine aminotransferase; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin.

According to the low value of laboratory reference values of $CD4^+T$ count, the 395 patients were divided into two groups: lower $CD4^+T$ group and higher $CD4^+T$ group. Patients in the lower group were older (55.0 ± 16.5 vs 51.3 ± 14.8, $P = 0.033$), contained more males (115/195 [59.0%] vs 89/111 [44.5%], $P = 0.004$), and more likely to have shortness of breath (75/195 [38.5%] vs 43/200 [21.5%], $P < 0.001$) and fever on admission (141/195 [72.3%] vs 122/200 [61.0%], $P = 0.017$). And, there were no significant difference in the proportion of comorbidities, including hypertension, diabetes, coronary heart disease, hepatitis of B infection and chronic obstructive pulmonary disease, between the two groups. Analysis of moderate and severe patients alone, also showed the same trend (Supplementary table1, Supplementary table 2).

3.2 Laboratory And Radiographic Findings

Of these 395 patients, median (IQR) values of Hs-CRP (5.0 [2.2–22.9] mg/L) and PCT (0.05 [0.03–0.08] ng/ml) were elevated, while the median (IQR) values of lymphocyte count, $CD4^+T$ count, $CD8^+T$ count were within standard ranges (Table 1). In moderate patients, only the median (IQR) value of Hs-CRP was elevated. (Supplementary table1.). In severe patients, median (IQR) values of Hs-CRP, PCT and D-dimer were elevated, while the median (IQR) values of lymphocyte count, $CD4^+T$ count, $CD8^+T$ count were decreased. (Supplementary table2). According to lung CT (computed tomography, CT) findings, in all patients, the proportion of ground-glass opacity and local patchy shadowing was 48.7% (170/349) and 38.7% (135/349), respectively.

In terms of laboratory findings, compared with patients in higher $CD4^+$T group, patients in the lower $CD4^+$T group showed lower median lymphocyte count (0.8 (0.6-1.0) vs 1.5 (1.2–1.8), $P < 0.001$, cells × 10^9/L), $CD8^+$T count (168.0 [107.0-250.0] vs 322.0 [244.3-443.5], $P < 0.001$, cells/ul), CD4/CD8 (1.4 [1.1–1.9] vs 1.8 [1.4–2.3], $P < 0.001$), but a higher median hypersensitive C-reactive protein (Hs-CRP) (8.2 [5.0-48.5] vs 4.9 [1.1-7.0], $P < 0.001$, mg/L) and procalcitonin (PCT) (0.06 [0.04–0.11 vs 0.04 [0.02–0.06], $P < 0.001$, ng/ml) (Table 1). Analysis of moderate and severe patients alone showed that lymphocyte count and $CD8^+$T were more commonly reduced in severe patients. There was no significant change in the proportion of $CD4^+$T lower than the lower limit of normal in moderate and severe patients, but the proportion of $CD4^+$T lower than the lower limit of normal in moderate and severe patients accounted for 48.2% (95/197) and 50.5% (100/198), respectively. (Fig. 2A). The analysis also found that it is the $CD8^+$T count that reflects the severity of the patient's condition, not the $CD4^+$T count. (Fig. 2B).

In terms of computed tomography findings, in moderate patients, compared with patients in the higher group, patients in the lower group more often represented as local patchy shadowing (45 [47.4%] vs 33 [32.4%], $P = 0.031$). Ground-glass opacity and local patchy shadowing did not differ between the two groups in the entire patient population. (Table 1).

3.3 Treatment And Clinical Outcome

In all cases, the proportion of use of oxygen inhalation, and mechanical ventilation were 84.3% (328/389), and 7.7% (30/388), respectively. The most common therapy is treatment with antiviral treatment (388/395, 98.2%), followed by antibiotic treatment (179/395, 45.3%), glucocorticoids treatment (94/395, 23.8%), intravenous immunoglobulin treatment (71/395, 18.2%), and only four patients (4/395, 1.0%) were treated with antifungal drugs. During follow-up, 27 patients died (27/395, 6.8%), and the rest were discharged (368/395, 93.2%).

Compared with patients in the higher $CD4^+$T group, patients in the lower group needed more oxygen inhalation (174/193 ,90.2% vs 154/196, 78.6%, $P = 0.002$), mechanical ventilation (26/193, 13.5% vs 4/195, 2.1%, $P < 0.001$), antibiotic treatment (112/195, 57.4% vs 67/200, 33.5%, $P < 0.001$) and glucocorticoids treatment (64/195, 32.8% vs 30/200, 15.0%, $P < 0.001$). Other treatments were similar between the two groups, such as antiviral treatment, intravenous immunoglobulin treatment, and antifungal treatment. The case in-hospital death rate was significant higher in patients with lower $CD4^+$T level than in those with higher $CD4^+$T level (25/195, 12.8% vs 2/200, 1.0%, $P < 0.001$). The detailed treatment of moderate and severe patients were shown in supplementary table1 and supplementary table2.

3.4 Survival Curves Of In-hospital Death

Kaplan-Meier survival curves of the COVID-19 patients grouped by $CD4^+$T count are shown in Fig. 3. The low $CD4^+$T group had a higher in-hospital death rate than the high $CD4^+$T group during the follow-up period (log rank < 0.001). The same trend was also found in severe patients (log rank < 0.001). Kaplan-Meier survival analysis was not performed on moderate patients because no patients died during follow-up.

3.5 Results of Cox proportional hazards analyses of in-hospital death

Cox proportional hazard regression analysis was performed to test the associations between the lower $CD4^+$T group and in-hospital death for COVID-19 patients. Results of univariate analyses indicated that patients with lower CD4 + T count exhibited a 13.659-fold increase in in-hospital death compared to patients with higher $CD4^+$T count (hazard ratio (HR) :13.659; 95% confidence intervals (CI):3.235–57.671). Meanwhile, age, history of hypertension, history of COPD, white blood cell count, lymphocyte count, $CD8^+$T lower group, required mechanical ventilation or glucocorticoids or intravenous immunoglobulin treatment or antibiotic treatment or antifungal treatment were correlated with the risk of in-hospital death in patients with severe COVID-19 (Supplementary Table 3).

Multivariate survival analysis was performed with Cox's proportional hazard regression model to identify the independent factors correlated with prognosis. After adjusting for age, sex and temperature (Mode 1), the HR of the lower $CD4^+$T group for in-hospital death was 14.182 (95%CI: 1.884-106.786, $P = 0.010$). After adjusting for history of hypertension, history of diabetes and shortness of breath (Mode 2), the HR of the lower $CD4^+$T group for in-hospital death was 13.631 (95%CI: 3.190-58.243, $P < 0.001$). After adjusting for white blood cells, platelet and creatinine (Mode 3), the HR of the lower $CD4^+$T group for in-hospital death was 8.170 (95%CI: 1.877–35.566, $P = 0.005$). After adjusting for hypersensitive C-reactive protein, procalcitonin and D-dimer (Mode4), the HR of the lower CD4 + T group for in-hospital death was 10.644 (95%CI: 2.439–46.458, $P = 0.002$). After adjusting for $CD8^+$T lower group and lymphocyte count lower group (Mode 5), the HR of the lower $CD4^+$T group for in-hospital death was 13.650 (95%CI: 1.976–94.279, $P = 0.008$). After adjusting for age, history of hypertension, shortness of breath, white blood cell count platelet count, D-dimer and CD4/CD8 (Mode 6), the HR of the low $CD4^+$T cells count group for in-hospital death was 7.656 (95%CI: 1.610-36.396, $P = 0.010$). Multivariate analysis demonstrated that presenting with lower $CD4^+$T count was an independent risk factor for in-hospital death. Variables like age, white blood cell count and shortness of breath also showed significance for independently predicting in-hospital death in this study (Table 2, Fig. 4). Similarly, Cox proportional hazards analyses was also performed on severe patients, and the results also suggested that lower $CD4^+$T count was an independent risk factor for in-hospital death (Supplementary table4, Supplementary table5, Supplementary Fig. 1).

Table 2
Results of multivariate Cox proportional-hazards regression analyzing the effect of baseline variables on in-hospital death in all patients.

Mode	HR (95%CI)	*P*
Not Adjusted $CD4^+T$, low vs. high	13.659 (3.235–57.671)	< 0.001
Mode 1		
$CD4^+T$, low vs. high	14.182 (1.884-106.786)	0.010
Sex, male vs. female	1.383 (0.561–3.406)	0.481
Age, per 1 year	1.093 (1.052–1.135)	< 0.001
Temperature, per 1℃	0.777 (0.445–1.354)	0.372
Mode 2		
$CD4^+T$, low vs. high	13.631 (3.190-58.243)	< 0.001
Hypertension, yes vs. no	5.823 (2.595–13.070)	< 0.001
Diabetes, yes vs. no	0.824 (0.322–2.113)	0.688
Shortness of breath, yes vs. no	7.848 (2.942–20.934)	< 0.001
Mode 3		
$CD4^+T$, low vs. high	8.170 (1.877–35.566)	0.005
WBC, per 1×10^9/L	1.294 (1.193–1.404)	< 0.001
PLT, per 1×10^9/L	0.992 (0.987–0.997)	0.003
Cr, per 1 umol/L	1.002 (0.995–1.009)	0.576
Mode 4		
$CD4^+T$, low vs. high	10.644 (2.439–46.458)	0.002
Hs-CRP, per 1 mg/L	0.989 (0.974–1.005)	0.193
PCT, per 1 ng/ml	1.017 (0.925–1.118)	0.724
D-dimer, per 1 mg/L	1.028 (1.018–1.038)	< 0.001
Mode 5		
$CD4^+T$, low vs. high	13.650 (1.976–94.279)	0.008

Abbreviations: WBC, white blood cell count; PLT, platelet count; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin; LYM, lymphocyte.

Mode	HR (95%CI)	*P*
$CD8^+T$, low vs. high	3.159 (0.853–11.707)	0.085
CD4/CD8 ratio, per 1 unit	1.422 (1.105–1.830)	0.006
LYM count, low vs. high	0.996 (0.306–3.243)	0.994
Mode 6		
$CD4^+T$, low vs. high	7.656 (1.610-36.396)	0.010
Age, per 1 year	1.074 (1.034–1.115)	< 0.001
Hypertension, yes vs. no	2.031 (0.766–5.386)	0.154
Shortness of breath, yes vs. no	3.435 (1.167–10.114)	0.025
WBC, per 1×10^9/L	1.224 (1.097–1.366)	< 0.001
PLT, per 1×10^9/L	0.996 (0.991–1.001)	0.149
D-dimer, per 1 mg/L	0.997 (0.992–1.002)	0.207
CD4/CD8 ratio, per 1 unit	1.106 (0.793–1.542)	0.552
Abbreviations: WBC, white blood cell count; PLT, platelet count; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin; LYM, lymphocyte.		

4. Discussion

This study showed the relationship between $CD4^+T$ count and in-hospital death in COVID-19 patients. The dominant symptoms observed in the study included fever on admission, cough, fatigue and shortness of breath. The most frequent comorbidities were hypertension and diabetes. Compared with patients with higher $CD4^+T$ level, patients with lower $CD4^+T$ level were older and were more frequently male. In terms of laboratory findings, lymphocyte count, $CD4^+T$ count, $CD8^+T$ count were significantly lower in lower group. The case in-hospital death rate was significant higher in patients with lower $CD4^+T$ level than in those with higher $CD4^+T$ level. After adjustment for potential confounding factors, the lower group remained a significant predictor for in-hospital death.

Previous studies have shown that $CD4^+T$ count was reduced significantly in COVID-19 patients [4]. It is suggested that $CD4^+T$ count and $CD8^+T$ count were reduced below the lower limit of normal in the vast majority of patients with either severe or moderate, and both of them were reduced profoundly in severe patients than in moderate patients [4]. In the present study, among 395 patients with COVID-19, 49.4% patients (195/395) showed decreased $CD4^+T$ count and the in-hospital death was markedly higher in patients with decreased $CD4^+T$ count than in patients with normal $CD4^+T$ count (12.8% vs 1.0%, P< 0.001). In addition, our study found that increased age and increased white blood cell count were

associated with in-hospital death, which were similar with several studies. Verity, et al. estimated that the total case fatality rate increased with age, with the case fatality rate of patients < 60 years old being 0.32% (95% CI: 0.27–0.38) and the case fatality rate of patients ≥ 60 years old being 6.4% (95% CI: 5.7–7.2), possibly because they often had other chronic diseases [8]. Wang, et al. suggested that white blood cell count and neutrophil count of dead patients were higher than those of surviving patients, which may be related to cytokine storm caused by the invasion of SARS-Cov-2 [9].

Several recent studies indicated that the absolute value of lymphocytes [1, 10] and T-lymphocytes [3, 11] were reduced in most patients with COVID-19. It was believed that SARS-CoV-2 may act mainly on lymphocytes, especially T-lymphocytes [4, 7]. At present, the potential mechanisms undergoing $CD4^+$T count decrease induced by SARS-CoV-2 infection is still unknown. Researchers analyzed the clinical characteristics of patients with COVID-19, consistently found that patients with COVID-19, especially those with severe COVID-19, had significantly higher concentrations of Interleukin-10 (IL-10), interferon-inducible protein 10 (IP10), monocyte chemo-attractant protein 1 (MCP-1/CCL2), macrophage inflammatory protein-1α (MIP1A/CCL3), tumor necrosis factor alpha (TNF-α) [2]. Meanwhile, it is reported that the concentration of IL-10, Interleukin-6 (IL-6), and TNF-α were negatively correlated with total T-cell counts, $CD4^+$T count, and $CD8^+$T count, respectively; Compared with patients in the illness period, levels of IL-10, IL-6, and TNF-α in the patients in the decline stage decreased significantly, while the total T-cell counts, $CD4^+$T count, and $CD8^+$T count were recovered [11]. The phenomena suggested the decrease in the number of T-cells in patients with COVID-19 may be due to the negative effects of high concentrations of TNF-α, IL-6, IL-10 in serum on the survival or proliferation of T-cells [11]. In addition, Previous studies have shown that, in SARS patients, the formation of autoimmune antibodies or immune complexes induced by viral infection and the use of steroids may play an important role in lymphocytic decline [12].

This study was limited by sample size and lack of dynamic detection of $CD4^+$T count and $CD8^+$T count. First, our study only analyzed 395 patients with COVID-19, the relatively small sample sizes may affect the statistical power. Secondly, the patients included in this study lacked dynamic measurements of $CD4^+$T count and $CD8^+$T count, which made the evaluation of the relationship between $CD4^+$T levels and disease changes in patients with COVID-19 incomplete.

5. Conclusions

In conclusion, the main findings of the study were that it is the $CD8^+$T count, not the $CD4^+$T count, that reflected the severity of the patient's disease; And, the high prognostic value of decreased $CD4^+$T count in patients with COVID-19. Lower CD^+4T count is independently associated with increased in-hospital death. Thus, in this acute-care setting, CD^+4T count can provide early prognostic information in patients with COVID-19.

Abbreviations

COPD, chronic obstructive pulmonary disease; CHD, coronary heart disease; SBP, systolic blood pressure; DBP, diastolic blood pressure; WBC, white blood cell count; Hb, Hemoglobin; PLT, platelet; LYM, lymphocyte; ALT, alanine aminotransferase; Cr, creatinine; Hs-CRP, hypersensitive C-reactive protein; PCT, procalcitonin.

Declarations

Ethics approval and consent to participate: All study procedures were approved by the local ethics committee (approval NO. 20200701). All subjects were well informed.

Consent for publication: All authors have read and approved the final manuscript.

Availability of data and materials: The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Competing interests: All authors declare no conflict of interest.

Funding: This work was supported by National Natural Science Foundation of China, 81970203; National Natural Science Foundation of China, 81570212; National Natural Science Foundation of China, 31800976; Chongqing Science and Health Joint Medical Research Project, 2018QNXM024.

Authors' Contributions: W.X. participated in study design, analyzing data analysis and manuscript writing. G.L., C.X., J.D., H.B., C.Y., L.P., and T.X. were involved in data collection. Q.S., C.G. and Z.D. were responsible for the study concept, design and final approval of manuscript. W.X. is the first author.

Acknowledgements: We thank all participants involved in this study. We thank all medical staff who participated in the fight against SARS-CoV-2.

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Figures

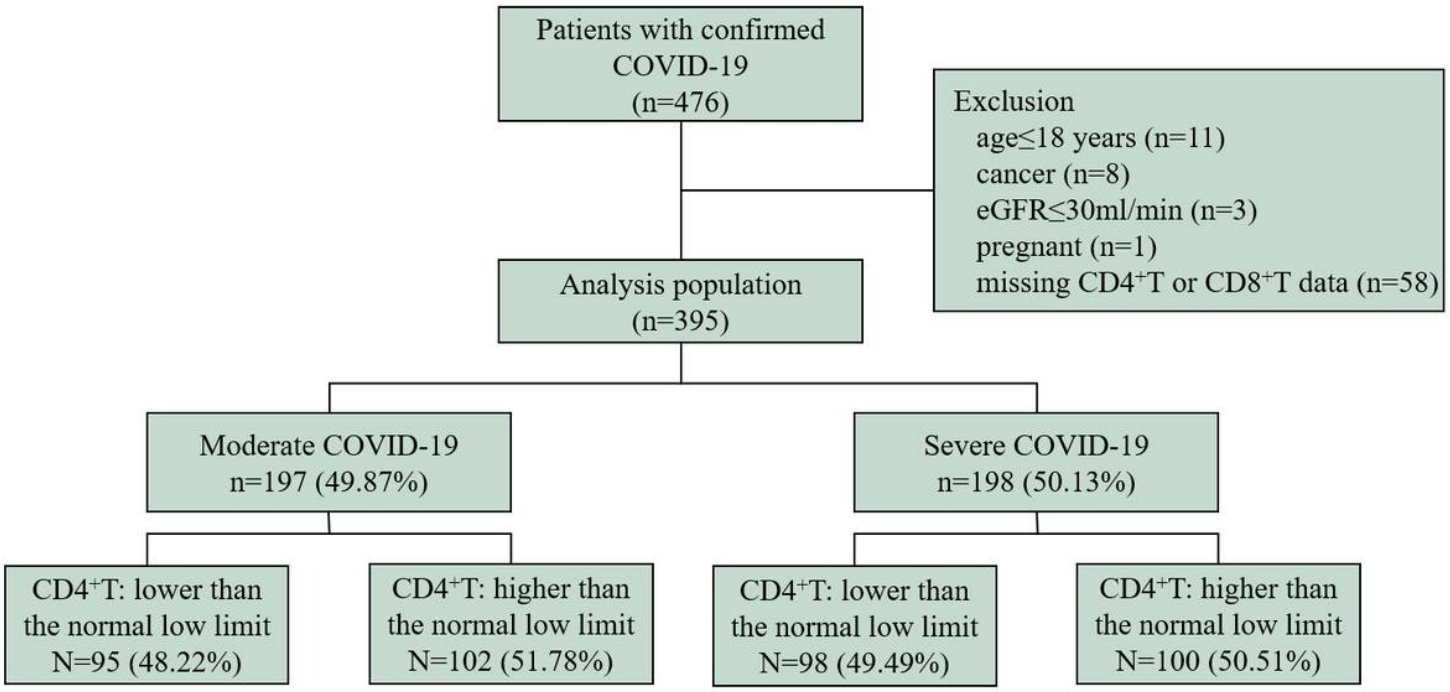

Figure 1

Flow diagram of Patient Recruitment

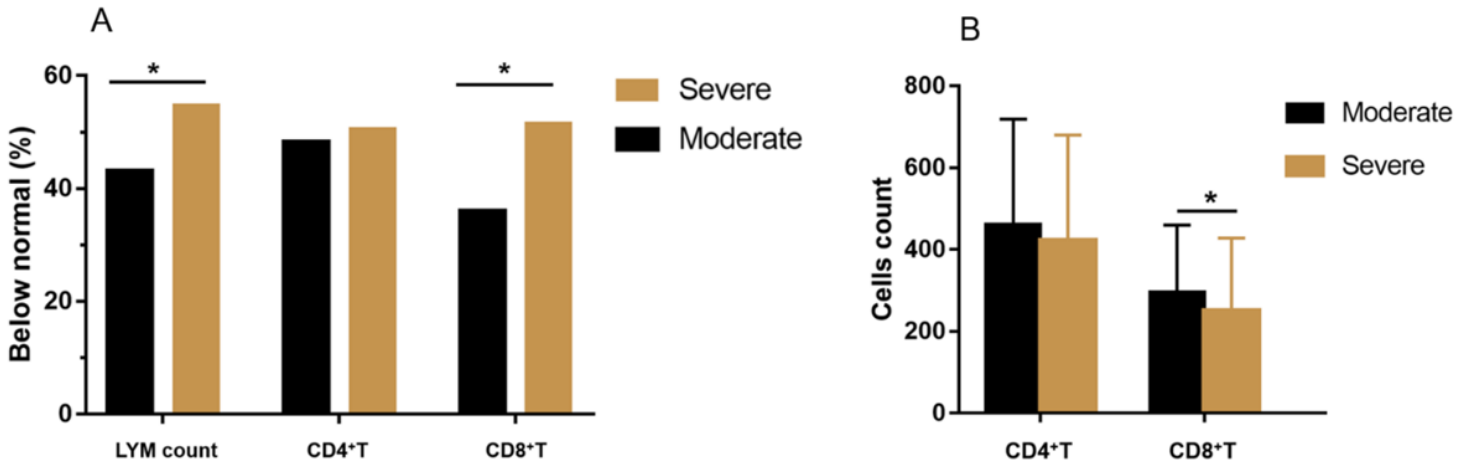

Figure 2

A: The histogram shows the proportion of moderate and severe patients with lymphocytes, CD4+T cells, and CD8+T cells below the lower limit of normal; B: The histogram shows the number of CD4+T cells and CD8+T cells in moderate and severe patients.

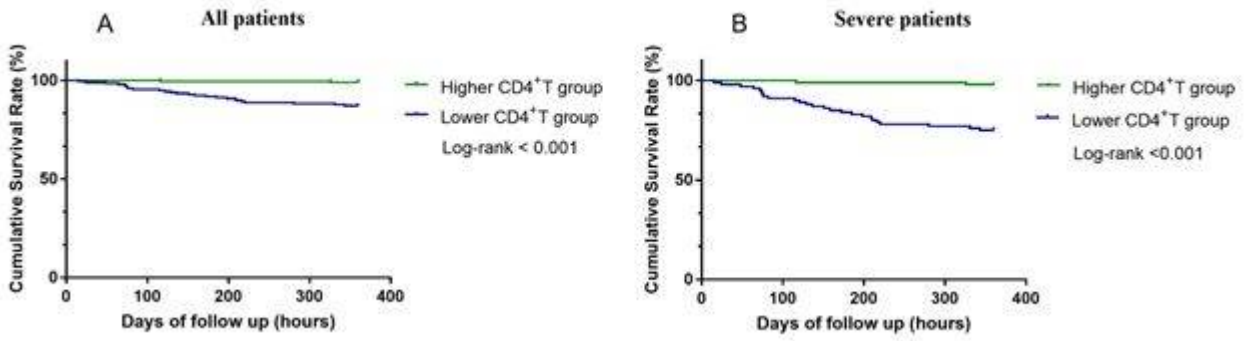

Figure 3

Kaplan-Meier plots showing the survival rate of COVID-19 patients who were stratified into two groups according to CD4+T cells count. (green line, higher CD4+T group; blue line, lower CD4+T group).

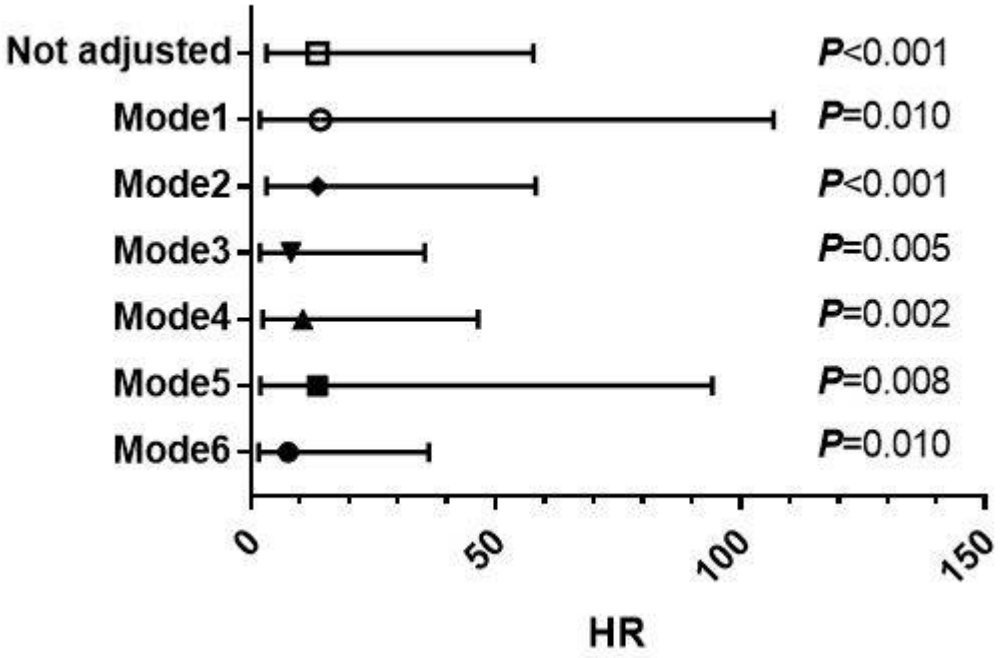

Figure 4

Forest plots of multivariate Cox proportional-hazards regression analyzing the effect of baseline variables on in-hospital death. Mode1: adjusted sex, age and temperature; Mode2: adjusted hypertension, diabetes and shortness of breath; Mode3: adjusted white blood cell count, platelet count and Creatinine; Mode4: adjusted hypersensitive C-reactive protein, procalcitonin and D-dimer; Mode5: adjusted CD8+T group, CD4/CD8 ratio and lymphocyte count group; Mode6: adjusted age, hypertension, shortness of breath, white blood cell count, platelet count, D-dimer and CD4/CD8 ratio.