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RESEARCH ARTICLE

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Risk factors for the COVID-19 severity and its correlation with viral shedding: A retrospective cohort study

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Abstract

Coronavirus disease 2019 (COVID-19) have become a pandemic in the world. This study is aim to explore risk factors for COVID-19 severity in the early stage and the correlation between the viral shedding and COVID-19 severity. We included inpatient with laboratory confirmed COVID-19 who had been discharged by 9 March 2020. The medical record data and dynamic change of biochemical indicators in-hospital were compared between common and severe patients. Eighty patients were included in this study. Multivariable regression demonstrated increasing odds of severity associated with the duration of fever (odds ratio [OR], 1.42; 95% confidence interval [CI], 1.10-1.82, per day increase; $P = .007$), C-reactive protein (CRP) (OR, 1.26; 95% CI, 1.04-1.52; $P = .02$), and $PO_2 < 80$ mm Hg (28.07, 95% CI, 1.50-524.12; $P = .026$) on admission. We found severe acute respiratory syndrome coronavirus 2 viral RNA could be long-term presence in respiratory tract and fecal sample, up to 43 and 46 days, respectively. However, the duration of viral shedding have no correlation with the COVID-19 severity. The duration of fever, elevated CRP and $PO_2 < 80$ mm Hg on admission were associated with the COVID-19 severity in the early stage and there is no correlation between the viral shedding and COVID-19 severity.

KEYWORDS

COVID-19, retrospective cohort study, risk factors, severity, viral shedding

1 | INTRODUCTION

Coronavirus disease 2019 (COVID-19), a disease caused by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2; previously known as 2019-nCoV), make a global pandemic with increasing number of COVID-19 cases confirmed around the world and result in alarmingly high mortality and morbidity.[1] As of 1 July 2020, cases have been reported in more than 200 countries, a total of more than 10.3 million cases in the global have been confirmed, and over 500 000 cases were death.[2]

The epidemiological features and clinical characteristics of COVID-19 have been previously reported.[3-6] The clinical spectrum of COVID-19 appears to be wide, ranging from asymptomatic infection to severe viral pneumonia with acute respiratory distress syndrome (ARDS) and even death. Recently, Zhou et al[7] first reported that older age, high SOFA score, and d-dimer greater than 1 μg/L on admission associated with increasing risk of in-hospital death in severe patients. Wu et al[8] showed that older age and fever were associated with greater risk of developing ARDS in patients with COVID-19.

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However, the estimation of risk factors for the severity of this disease in the early stage and the association between viral shedding and the severity of this disease is unclear. In this study, we investigate risk factors for the severity of patients with COVID-19 in the early stage and the relationship of viral shedding and the severity of COVID-19, and describe the dynamical changes of laboratory findings of inpatients from a single hospital in Zhuhai, China.

2 | METHODS

2.1 | Study design and participants

This study included two cohorts of inpatients from the Fifth Affiliated Hospital of Sun Yat-Sen University (Zhuhai, China), which was the only designated hospitals for transfer of patients with COVID-19 from other hospitals in Zhuhai city, Guangdong province. All patients diagnosed with COVID-19 according to WHO interim guidance were enrolled in our study, who were discharged between 17 January 2020 (ie, when the first patients were admitted) and 9 March 2020. Clinical classification for patients with COVID-19 according to the Chinese management guideline for COVID-19 (version 6.0)[9]: (a) mild type: clinical symptoms were mild, no pneumonia was found on imaging; (b) common type, with fever, respiratory tract and other symptoms, imaging evidence of pneumonia; (c) severe type: any of the following: shortness of breath, breathing frequency ≥30 times /min; At rest, oxygen saturation ≤93%; Arterial partial pressure (PaO_2)/oxygen absorption concentration (FiO_2) ≤300 mm Hg; (d) critical type: meet any of the following: respiratory failure, and the need for mechanical ventilation; Shock; or with other organ failure requiring intensive care unit (ICU) care. The early stage of patient with COVID-19 in our study was defined clinically as from the onset of viral infection to progression to common type COVID-19 (with fever, respiratory and other symptoms, and imaging changes of pneumonia) according to the clinical classification. To simplify the analysis process, the severe type and critical type cases diagnosed after discharge were combined as "severe type" in this study. Mild patients with no change in their condition during hospitalization were not included in this study. All patients included were divided into common and severe type group. Oral consent was obtained from patients. The clinical outcomes (ie, discharges, length of stay) were monitored up to 9 March 2020, the final date of follow-up. The study was approved by the Research Ethics Commission of the Fifth Affiliated Hospital of Sun Yat-Sen University (No·ZDWY[2020] Lunzi No·(K22-1)).

2.2 | Data collection

The medical records of patients were analyzed by the research team of Infectious Disease Prevention and Treatment center, the Fifth Affiliated Hospital of Sun Yat-Sen University. Epidemiological, clinical, laboratory, and treatment and outcomes data were collected with data collection forms from electronic medical records. These data were checked by a trained team of physicians. Information recorded included demographic data, medical history, exposure history, underlying comorbidities, symptoms, signs, laboratory findings, chest computed tomographic (CT) scans, and treatment measures (ie, antiviral therapy, corticosteroid therapy, antibiotics, and intravenous immunoglobin). The time of illness onset was defined as the day when the symptom was noticed. Fever was defined as axillary temperature of at least 37·3°C. The duration of fever was defined as from the day of fever symptom onset to the day of body temperature return to normal for three consecutive days after admission. Laboratory values every day and treatment measures during the hospital stay were also collected.

2.3 | Laboratory procedures

SARS-CoV-2 RNA in respiratory specimens was detected in our hospital lab by real-time reverse transcription-polymerase chain reaction and then was confirmed by the Chinese Center for Disease Control and Prevention, and only qualitative data were available. Throat-swab specimens were obtained for SARS-CoV-2 polymerase chain reaction re-examination every other day after clinical remission of symptoms. The criteria for discharge were absence of fever for at least 3 days, substantial improvement in both lungs in chest CT, clinical remission of respiratory symptoms, and two throat-swab and excrement samples negative for SARS-CoV-2 RNA obtained at least 24 hours apart. Routine blood examinations for blood count, coagulation profiles, serum biochemical tests (including renal and liver function, creatine kinase, and lactate dehydrogenase), myocardial enzymes tests, cardiac function detection arterial blood gas analysis were performed for all inpatients in our hospital. Frequency of examinations was determined by the treating physician.

2.4 | Statistical analysis

Continuous variables were described using mean ± standard deviation and categorical variables were presented as frequency rates and percentages. We use independent group t tests to compare means for continuous variables when the data were normally distributed; otherwise, the Mann-Whitney U test was used. For categorical variables, we used the χ^2 test, or Fisher's exact test to compare differences between common and severe type group where appropriate. To investigate the risk factors associated with the severity of COVID-19, univariable and multivariable logistic regression models were used. The correlation between the viral shedding of throat-swab and excrement and the severity of COVID-19 was determined by Kaplan-Meier method and positive ratio curves between different groups were calculated by log-rank test. A two-sided α of less than 0.05 was considered statistically significant. All statistical analyses were performed using SPSS 22.0 software (SPSS Inc).

3 | RESULTS

Eighty patients discharged from the Fifth Affiliated Hospital of Sun Yat-Sen University were enrolled to analyze in this retrospective

study. Twenty-eight patients were defined as severe type (six for ICU therapy, one of whom death) and 52 for common type according to the Chinese management guideline for COVID-19 (version 6.0). The mean age of the 80 patients was 50.2 years, ranging from 15 years to 80 years. The age of severe patients with COVID-19 was markedly older than that in the common patients (Table 1). 66 (82.5%) patients have Wuhan city or Hubei province exposure history. Comorbidities were present in half of patients, and hypertension (17 [21.3%]) was the most common comorbidity, followed by diabetes (8 [10%]), cardiovascular disease (4 [5%]) and carcinoma (4 [5%]) (Table 1). The most common symptoms on admission were fever (48 [60%]), followed by dry cough (33 [41.3%]), expectoration (15 [18.8%]), and sore throat (14 [17.5%]) (Table 1). The mean time of hospital stays was 20.3 days (20.3 ± 7.0), and the mean time from illness onset to hospital admission was 4.8 days (4.8 ± 4.9). The duration of fever was significant difference between common and severe type patients (2.4 ± 3.8 vs 7.1 ± 5.2 days; $P = .000$). The temperature on admission in severe patient was markedly higher than that in common patients (37.4 ± 0.6 vs 36.9 ± 0.6). There was no difference in blood type between the two groups.

For laboratory findings, we found that lymphocyte count, lymphocyte percentage, platelet count, eosinophil count, albumin, oxygen partial pressure (PO_2) and partial pressure of carbon dioxide (PCO_2) in severe patients significantly decreased on admission when compared with common patients, otherwise, aspartate aminotransferase (AST), lactate dehydrogenase (LDH), Alpha-hydroxybutyric dehydrogenase, C-reactive protein (CRP), D-dimer, and fibrinogen showed increase in severe patients (Table 2). In regard to therapy, 76 (95%) patients received antivirals (lopinavir/ritonavir, arbidol and chloroquine phosphate). Antibacterial drugs and antifungal agents were used more frequent in severe patients compared with common patients (Table 3). Systematic corticosteroid, intravenous immunoglobulin, fresh frozen plasma and human serum albumin use differed significantly between common and severe patients (Table 3). 100% patients in the severe patients were treated with oxygen inhalation and six (7.5%) patients were transferred to ICU treatment (Table 3).

In univariable analysis, we found that age, temperature on admission, the duration of fever, lymphopenia, platelet count, reduced albumin, direct bilirubin, elevated AST, LDH, Alpha-hydroxybutyric dehydrogenase, CRP, d-dimer, fibrinogen, oxygen partial pressure, and partial pressure of carbon dioxide were associated with the severity of COVID-19 (Table 4). Then we analyzed 80 patients with complete data for all variables in the multivariable logistic regression model. We found that the duration of fever (odds ratio [OR], 1.42; 95% confidence interval [CI], 1.10-1.82, per day increase; $P = .007$), CRP (OR, 1.26; 95% CI, 1.04-1.52; $P = .02$), oxygen pressure less than 80mm Hg (OR, 28.07; 95% CI, 1.50-524.12; $P = .026$) on admission were associated with increased odds of the severity of COVID-19 (Table 4).

To explore the dynamic change of biochemical indicators in inpatients with COVID-19, we recorded the result data of laboratory tests performed daily in the hospital from the onset of illness to discharge. We found that within 20 days of illness onset, lymphocyte count, lymphocyte percentage, and platelet count were lower in the severe patients than in the normal patients, and these indicators tended to overlap after treatment intervention (Figure 1A-C). Interestingly, we found that patients with COVID-19 showed a gradual increase in body temperature and CRP within 1 week of illness onset, and a gradual decrease and normalcy after treatment intervention (Figure 1D,E). In the early stage of the disease (about 2 weeks), the body temperature and CRP of patients in the severe patients were higher than those in the common patients (Figure 1D,E). In addition, we also observed that patients with severe illness are more likely to have increased N-terminal pro-brain natriuretic peptide (NT-proBNP) in the course of disease (Figure 1F), suggesting that these patients are prone to cardiac dysfunction. Then we observed the dynamical change of the markers of other routine blood test, liver function, myocardial enzyme, blood coagulation function and renal function, we find that these indicators do not show obvious difference as a whole in both group patients (Figure S1-4). Interestingly, the red blood cell and hemoglobin decreased gradually but eosinophils increased gradually in both common and severe patients within 24 days from illness onset (Figure S1C-F). During hospitalization time, a few severe patients showed elevated white blood cells and neutrophils (Figure S1A,B), anemia (Figure S1C,D), elevated bilirubin (Figure S2C,D), an increase of CK, lactate dehydrogenase and alpha hydroxyl butyric acid dehydrogenase (Figure S3A, C, and D) and elevated D-dimer (Figure S4E).

To explore the correlation between the viral shedding and the disease severity, we compared the viral RNA positive ratio of throat-swab and excrement sample between common and severe patients (Figure 2). The longest observed duration of viral shedding for throat-swab and fecal sample in patients with COVID-19 was 43 and 46 days after illness onset, respectively. There is no difference for SARS-CoV-2 RNA positive ratio in throat-swab (Figure 2A) and excrement (Figure 2B) sample between common and severe patients, suggesting there is no correlation between the duration of viral shedding and the severity of COVID-19.

4 | DISCUSSION

In this study, we found the duration of fever, CRP and PO_2 < 80mm Hg on admission were associated with higher risk of the severity of patients with COVID-19 in the early stage of disease and there is no correlation between the duration of viral shedding and the severity of COVID-19. We also found that lymphocyte, lymphocyte percentage, and platelet within 20 days of illness onset were lower in the severe patients compared with common patients, the red blood cell and hemoglobin decreased gradually but eosinophils increased gradually in patients with COVID-19 within 24 days from illness onset. Additionally, elevated levels of AST, bilirubin, NT-proBNP, D-dimer, CK, LDH, and lymphopenia were more commonly observed in hospital in severe patients with COVID-19.

A total of 80 patients with COVID-19 were included in this study, most of whom had a exposure history of Wuhan city or Hubei province. Consistent with previous reports on clinical

TABLE 1 Demographics and clinical characteristics of patients with COVID-19

	Total (n = 80)	Severe (n = 28)	Common (n = 52)	*P* value
Age, y	50.2 ± 15.1	56.3 ± 14.0	46.9 ± 14.8	.007
Sex				.052
Male	34/80 (42.5%)	16/28 (57.1%)	18/52 (34.6%)	
Female	46/80 (57.5%)	12/28 (42.9%)	34/52 (65.4%)	
Wuhan or Hubei exposure history	66/80 (82.5%)	21/28 (75%)	45/52 (86.5%)	.226
Comorbidity	40/80 (50%)	16/28 (57.1%)	24/52 (46.2%)	.348
Hypertension	17/80 (21.3%)	10/28 (35.7%)	7/52 (13.5%)	.020
Cardiovascular disease	4/80 (5%)	2/28 (7.1%)	2/52 (3.8%)	.609
Diabetes	8/80 (10%)	6/28 (21.4%)	2/52 (3.8%)	.019
Carcinoma	4/80 (5%)	1/28 (3.6%)	3/52 (5.8%)	1.000
Cerebrovascular Disease	2/80 (2.5%)	1/28 (3.6%)	1/52 (1.9%)	1.000
Chronic kidney disease	1/80 (1.3%)	1/28 (3.6%)	0	.350
Chronic lung disease	3/80 (3.8%)	2/28 (7.1%)	1/52 (1.9%)	.279
Other	23/80 (28.8%)	7/28 (25.0%)	16/52 (30.8%)	.587
Symptoms & signs				
Fever (temperature ≥37.3℃)	48/80 (60%)	23/28 (82.1%)	25/52 (48.1%)	.003
Fatigue	8/80 (10%)	6/28 (21.4%)	2/52 (3.8%)	.019
Myalgia	10/80 (12.5%)	6/28 (21.4%)	4/52 (7.7%)	·090
Headache	7/80 (8.8%)	5/28 (17.9%)	2/52 (3.8%)	.048
Dizziness	4/80 (5%)	1/28 (3.6%)	3/52 (5.8%)	1.000
Sore throat	14/80 (17.5%)	6/28 (21.4%)	8/52 (15.4%)	.497
Nasal obstruction	4/80 (5%)	2/28 (7.1%)	2/52 (3.8%)	.609
Snot	5/80 (6.3%)	2/28 (7.1%)	3/52 (5.8%)	1.000
Dry cough	33/80 (41.3%)	11/28 (39.3%)	22/52 (42.3%)	.817
Expectoration	15/80 (17.3%)	6/28 (21.4%)	9/52 (17.3%)	.652
Dyspnea	5/80 (6.3%)	4/28 (14.3%)	1/52 (1.9%)	.048
Diarrhea	5/80 (6.3%)	3/28 (10.7%)	2/52 (3.8%)	.337
Nausea or vomiting	3/80 (3.8%)	0	3/52 (5.8%)	.548
Time from illness onset to Hospital admission, days	4.8 ± 4.9	4.4 ± 3.4	5.0 ± 5.6	·627
Hospital stays, days	20.3 ± 7.0	20.3 ± 6.0	20.2 ± 7.5	·947
Duration of fever, days	4.0 ± 5.8	7.1 ± 5.2	2.4 ± 3.8	.000
Temperature (℃) on admission	37.1 ± 0.6	37·4 ± 0·6	36.9 ± 0·6	.000
Pulse, beats per min	86.0 ± 13.7	86.0 ± 14.3	86.0 ± 13.6	.986
Respiratory rate, breaths per min	19.3 ± 2.0	19.2 ± 2.2	19.4 ± 1.9	.616
Systolic blood pressure, mm Hg	131.7 ± 17.5	130.9 ± 19.0	132.2 ± 16.9	.776
Diastolic blood pressure, mm Hg	84.3 ± 10.4	83.4 ± 11.3	84.8 ± 10.0	.190
Blood type[a]				.081
A	26/69 (37.7%)	5/25 (20%)	21/44 (47.7%)	
B	14/69 (20.3%)	5/25 (20%)	9/44 (20.5%)	
AB	10/69 (14.5%)	6/25(24%)	4/44 (9.1%)	
O	19/69 (27.5%)	9/25 (36%)	10/44 (22.7%)	

Note: Data are mean ± SD, or n/N (%). *P* values were calculated by the *t* test, χ^2 test, or Fisher's exact test, as appropriate.
Abbreviation: COVID-19, coronavirus disease 2019.
[a]χ^2 test comparing all subcategories.

characteristics,[4,5,10,11] the common symptoms of patients with COVID-19 in this study were fever, dry cough, sputum cough, sore throat, fatigue, and some atypical symptoms, such as diarrhea, vomiting, nausea, headache, and nasal congestion. Common complications include hypertension, diabetes, and cardiovascular disease. In this study, the age, proportion of hypertension and diabetes and the duration of fever of severe patients were greater than that of common patients with COVID-19, suggesting that age,

TABLE 2 Laboratory findings of patients with COVID-19 on admission to hospital

Parameters	Normal range	Total (n = 80)	Severe (n = 28)	Common (n = 52)	*P* value
White blood cell count, $\times10^9$/L	3.5-9.5	5.1 ± 1.8	4.9 ± 1.6	5.2 ± 1.8	.428
Red blood cell count, $\times10^{12}$/L	4.3-5.8	4.5 ± 0.5	4.6 ± 0.7	4.5 ± 0.5	.395
Neutrophil count, $\times10^9$/L	1.8-6.3	2.9 ± 1.5	3.1 ± 1.7	2.8 ± 1.4	.432
Lymphocyte count, $\times10^9$/L	1.1-3.2	1.5 ± 0.7	1.2 ± 0.4	1.7 ± 0.7	.001
Lymphocyte percentage, %	20-50	31.7 ± 10.7	27.5 ± 10.1	34.0 ± 10.4	.008
Platelet count, $\times10^9$/L	125-350	194.8 ± 65.3	156·7 ± 42.4	215.3 ± 66.4	.000
Monocyte count, $\times10^9$/L	0.1-0.6	0.5 ± 0.2	0.5 ± 0.2	0.5 ± 0.2	.886
Eosinophil count, $\times10^9$/L	0.02-0.52	0.04 ± 0.06	0.02 ± 0.03	0.05 ± 0.07	.028
Hemoglobin, g/L	130-175	136.7 ± 17.7	139.3 ± 19.9	135.3 ± 16.5	.339
ALT, U/L	9-50	21.85 ± 15.3	25.1 ± 16.3	20.1 ± 14.6	.170
AST, U/L	15-40	22.3 ± 8.9	26.2 ± 10.6	20.2 ± 7.1	·003
Total bilirubin, μmol/L	3-24	8.6 ± 4.0	9.3 ± 4.4	8.2 ± 3.7	.216
Direct bilirubin, μmol/L	0-8	3.4 ± 1.5	3.9 ± 1.7	3.2 ± 1.3	.028
Total protein, g/L	65-85	69.2 ± 5.1	67.9 ± 5.2	69.9 ± 5.0	.096
Albumin, g/L	40-55	39.3 ± 3.5	37.6 ± 3.2	40.2 ± 3.3	.001
LDH, U/L	120-250	176.7 ± 47.2	197.2 ± 50.9	165.7 ± 41.4	.004
Alpha-hydroxybutyric dehydrogenase, U/L	72-182	138.3 ± 33.3	153.0 ± 36.2	130.4 ± 28.9	.003
Creatine kinase, U/L	26-192	88.7 ± 105.6	111.1 ± 165.3	76.7 ± 48.7	.166
CK-MB, U/L	0-25	13.1 ± 4.2	13.4 ± 4.0	13.0 ± 4.3	.671
CRP, mg/L	0.068-8.2	13.5 ± 23.8	28.2 ± 33.8	4.98 ± 5.30	·000
Urea, mmol/L	3.1-8	4.0 ± 2.6	4.6 ± 3.9	3.6 ± 1.5	.114
Creatinine, μmol/L	57-111	65.9 ± 38.3	76.7 ± 57.9	60.1 ± 20.0	.064
NT-proBNP, pg/mL	0-125	138.1 ± 194.7	194.8 ± 242.0	91.9 ± 133.3	.065
Prothrombin time, s	9.4-12.5	12.1 ± 1.1	12.5 ± 1.2	12.0 ± 1.0	.065
APTT, s	25.1-36.5	30.9 ± 3.4	31.3 ± 3.9	30.7 ± 3.1	.466
Thrombin time, s	10.3-16.6	13.7 ± 1.7	13.5 ± 1.3	13.9 ± 1.9	.249
Fibrinogen, g/L	2.38-4.98	3.1 ± 0.9	3.6 ± 0.8	2.9 ± 0.8	.001
D-dimer, ng/mL	0-243	124.4 ± 115.2	165.8 ± 128.1	102.1 ± 102.1	.017
PO_2, mm Hg	83-108	95.2 ± 25.6	83.9 ± 13.7	101.4 ± 28.4	.003
<80		15/79 (19%)	10/28 (35.7%)	5/51 (9.8%)	·005
≥80		64/79 (81%)	18/28 (64.3%)	46/51 (90.2%)	...
Whole blood lactic acid, mmol/L	0.5-1.6	1.6 ± 0.6	1.5 ± 0.7	1.6 ± 0.6	.710
PCO_2, mm Hg	35-45	38.9 ± 4.2	37.4 ± 4.5	39.9 ± 3.8	.010
pH value	7.35-7.45	7.398 ± 0.033	74.6 ± 0.037	7.394 ± 0.031	.151

Note: Data are mean ± SD, or n/N (%). *P* values were calculated by the *t* test, χ^2 test, or Fisher's exact test, as appropriate.
Abbreviations: ALT, alanine aminotransferase; APTT, activated partial thromboplastin time; AST, aspartate aminotransferase; CK-MB, creatine kinase muscle-brain isoform; COVID-19, coronavirus disease 2019; CRP, C-reactive protein; LDH, lactate dehydrogenase; NT-proBNP, N-terminal pro-brain natriuretic peptide; PO_2, oxygen partial pressure; PCO_2, partial pressure of carbon dioxide.

hypertension, diabetes and the duration of fever may be risk factors for poor prognosis of patients, and recent literature studies indicate that elder age and high fever were associated with higher risk of developing ARDS in the patients with COVID-19.[12] Many studies have analyzed risk factors for death and severe complications in patients with severe COVID-19.[7,12,13] To the best of our knowledge, there is no related study about risk factors associated with COVID-19 illness severity. This study using multiple factors regression

TABLE 3 Treatments of patients with COVID-19

	Total (n = 80)	Severe (n = 28)	Common (n = 52)	*P* value
Antiviral treatment	76/80 (95%)	26/28 (92.9%)	50/52 (96.2%)	.609
Antibacterial drugs	51/80 (63.8%)	28/28 (100%)	23/52 (44.2%)	·000
Antifungal agents	9/80 (11.3%)	8/28 (28.6%)	1/52 (1.9%)	.001
Corticosteroids	21/80 (26.3%)	15/28 (53.6%)	6/52 (11.5%)	.000
Intravenous immunoglobin	56/80 (70%)	27/28 (96.4%)	29/52 (55.8%)	.000
Fresh frozen plasma	26/80 (32.5%)	21/28 (75.0%)	5/52 (9.6%)	·000
Thymalfasin	29/80 (36.3%)	13/28 (46.4%)	16/52 (30.8%)	.165
Human serum albumin	51/80 (63.8%)	26/28 (92.9%)	25/52 (48.1%)	·000
Oxygen inhalation	47/80 (58.8%)	28/28 (100%)	19/52 (36.5%)	.000
ICU therapy	6/80 (7.5%)	6/28 (21.4%)	0	.01

Note: Data are mean ± SD, or n/N (%). *P* values were calculated by the *t* test, χ^2 test, or Fisher's exact test, as appropriate.
Abbreviations: COVID-19, coronavirus disease 2019; ICU, intensive care unit.

analysis shows that elevated CRP, oxygen partial pressure less than 80mm Hg and the duration of fever were associated with the severity of disease; which may be helpful to clinicians to judge the disease severity and to actively make a treatment avoiding illness aggravation and provide a reference for evaluating therapeutic effect at the same time.

In this study, the lymphocyte count, lymphocyte percentage and platelet count of severe patients on admission were significantly

TABLE 4 Risk factors associated with the severity of patients with COVID-19

	Univariable OR (95% CI)	*P* value	Multivariable OR (95% CI)	*P* value
Age, y	1.05 (1.01-1.08)	.010		
Sex				
Male	0.40 (0.16-1.02)	.055		
Female	1 (ref)			
Temperature on admission, ℃	4.26 (1.78-10.21)	.001		
Duration of fever, d	1.27 (1.11-1.44)	·000	1.42 (1.10-1.82)	.007
Lymphocyte count, $\times 10^9$/L	0.19 (0.07-0.57)	.003		
Lymphocyte percentage	0.94 (0.89-0.99)	.012		
Platelet count, $\times 10^9$/L	0.98 (0.97-0.99)	.001		
Albumin, g/L	0.776 (0.66-0.92)	.003		
Direct bilirubin, μmol/L	1.42 (1.03-1.96)	.033		
AST, U/L	1.09(1.02-1.15)	.007		
LDH, U/L	1.02 (1.01-1.03)	.008	1.03 (0.99-1.07)	.056
Alpha-hydroxybutyric dehydrogenase, U/L	1.02 (1.01-1.04)	.006		
C-reactive protein, mg/L	1.17 (1.08-1.28)	.000	1.26 (1.04-1.52)	.02
D-dimer, ng/ml	1.01 (1.00-1.01)	.031	0.99 (0.98-1.00)	.048
Fibrinogen, g/L	2.90 (1.44-5.82)	.003		
PO_2, mm Hg				
<80	5.11 (1.53-17.4)	.008	28.07 (1.50-524.12)	.026
≥80	1 (ref)		1 (ref)	
PCO_2, mm Hg	0.86 (0.76-0.97)	.014		

Abbreviations: AST, aspartate aminotransferase; CI, confidence interval; COVID-19, coronavirus disease 2019; LDH, lactate dehydrogenase; OR, odds ratio; PO_2, oxygen partial pressure; PCO_2, partial pressure of carbon dioxide.

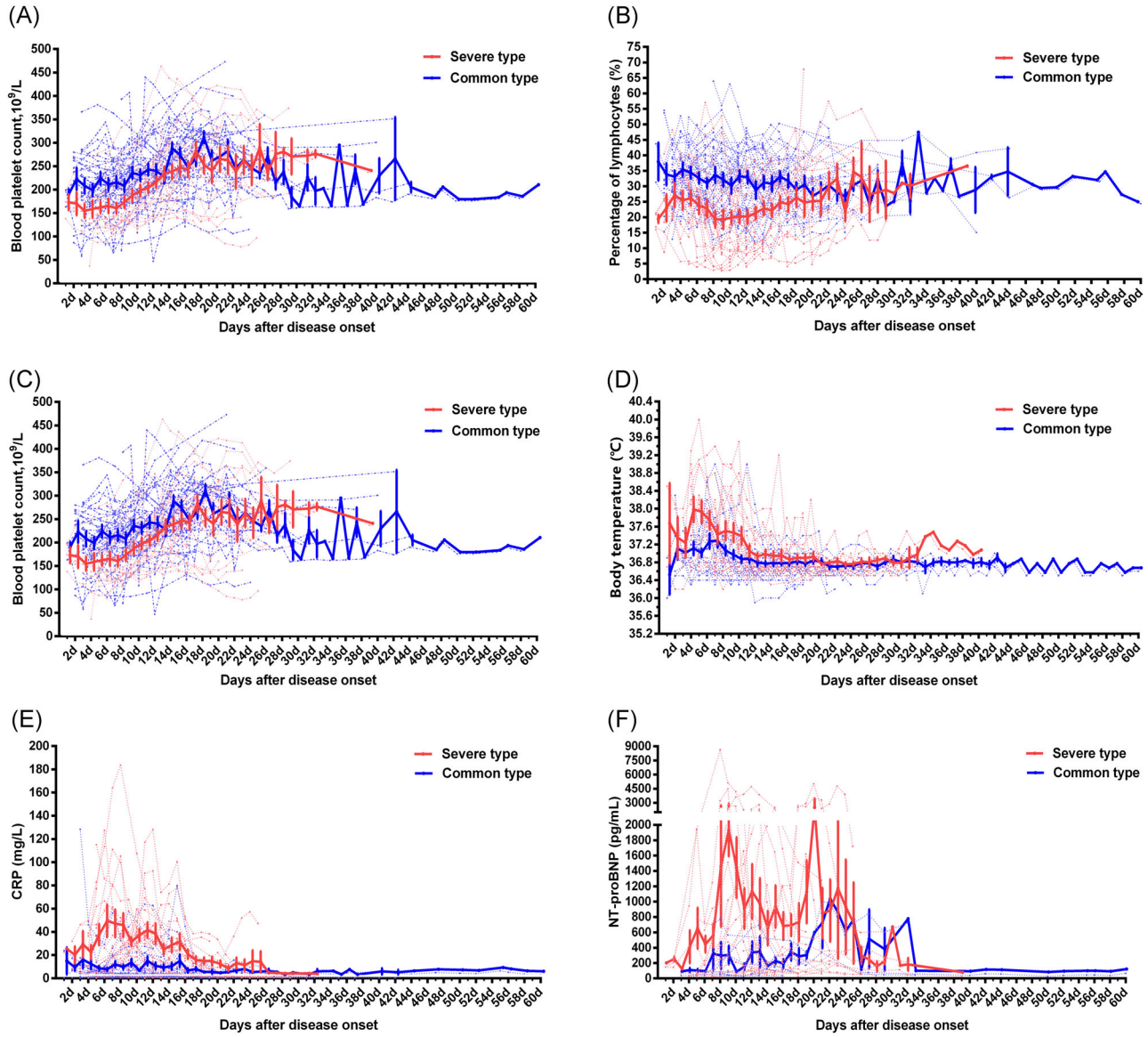

FIGURE 1 Dynamical changes in laboratory findings from illness onset in patients hospitalized with COVID-19. Figure shows dynamical changes in (A) lymphocytes, (B) lymphocytes percentage, (C) blood platelet, (D) body temperature, (E) CRP, (F) and NT-proBNP. Data was showed by mean ± SEM. COVID-19,coronavirus disease 2019; CRP, C-reactive protein; NT-proBNP, N-terminal pro-brain natriuretic peptide

lower than those of common type patients. Dynamic observation of these experimental indicators also decreased within 20 days of illness onset and gradually returned to normal after treatment in severe patients. And no significant changes were present in white blood cells and neutrophils between severe and common patients. The biopsy of patient died of COVID-19 revealed a large number of lymphocyte infiltration in the lung.[14] Studies have demonstrated that ACE2 receptor is highly expressed in lung tissue, which is the binding receptor of SARS-CoV-2 invasion in human body.[15,16] These data at present may indicate that SARS-CoV-2 firstly cause a cellular immune response in the lungs in the early infection and lead to plenty of peripheral blood lymphocyte migration into the lung causing the decrease of peripheral blood lymphocytes and pulmonary interstitial lesions radiographically and the lymphocyte infiltration of lung pathologically, and no obvious inflammation manifestation, such as elevated white blood cells, lung exudative lesions in early stage. Therefore, more basic research is needed to explore the pathogenesis of COVID-19.

Respiratory transmission is the primary route for SARS-CoV-2 and there is evidence for gastrointestinal infection of SARS-CoV-2.[17] Consistent with previous reports,[7,18] in our study we found SARS-CoV-2 viral RNA could be long-term presence in respiratory tract and fecal sample, up to 43 and 46 days, respectively. Previous studies have reported that viral RNA copy numbers is positively associated with older age, but not with disease severity.[19] Fu et al[20] have shown that CHD comorbidity, decreased albumin levels and delayed antiviral therapy experienced delays in clearance of SARS-CoV-2 RNA. Studies also have shown that age, male sex, delayed admission, and

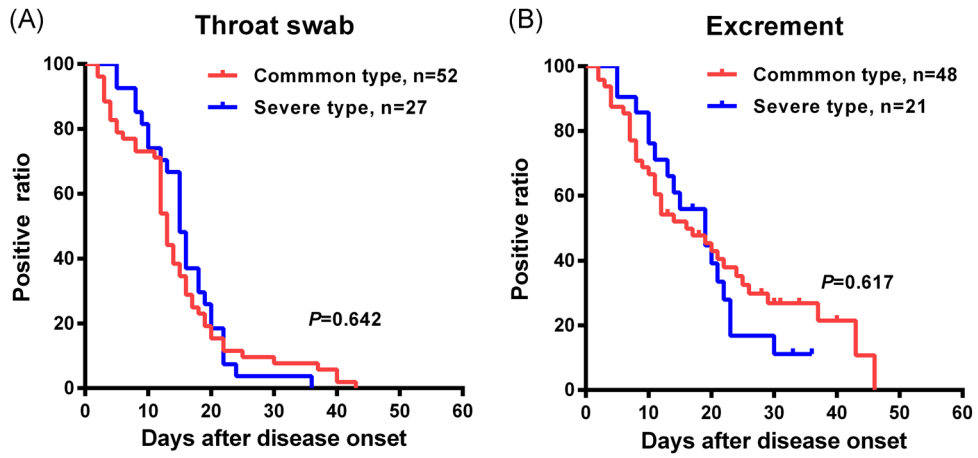

FIGURE 2 SARS-CoV-2 viral RNA positive ratio of common and severe patient with COVID-19. Figure show the SARS-CoV-2 viral RNA positive ratio in throat-swab sample (A) and excrement sample (B) in common and severe patient with COVID-19. COVID-19, coronavirus disease 2019. SARS-CoV-2, severe acute respiratory syndrome coronavirus 2

invasive mechanical ventilation are independent risk factors for prolonged viral shedding.[21,22] However, our study showed that the viral RNA clearance in throat-swab and fecal specimens have no correlation with the severity of the disease. These results suggest that viral shedding of SARS-CoV-2 may be primarily determined by the host's own immune state, so in-depth study of the interaction between SARS-CoV-2 and the host's immune system is needed, which could provide guidance for assanation and identify an appropriate treatment for viral RNA clearance.

Cardiac complications, including acute heart failure, arrhythmia, and myocardial infarction, are more common in patients with pneumonia. Cardiac arrest happened in about 3% pneumonia inpatients.[23] Older age, the severity of pneumonia and pre-existing cardiovascular disease were associated with higher risk for cardiac events in pneumonia patients.[24] Similarly, older and hypertensive patients with COVID-19 were more likely to be severely ill in our study. Previous studies reported that patients with COVID-19 have higher levels of cardiac enzymes and myocardial necrosis markers.[3,4,25] Consistent with these results, severe patients with COVID-19 in our study easily occurred an increase of NT-proBNP, CK, LDH andα-hydroxybutyrate dehydrogenase, indicating the presence of cardiac dysfunction and cardiac injury. Based on the invasion of SARS-CoV-2 to cell through binding of ACE2 receptor and cardiac tissue expressing ACE2,[15] the mechanism of cardiac injury related with COVID-19 was theoretically present that SARS-CoV-2 may directly damage the heart by ACE2 receptor. Therefore ACE2 related signaling pathway may play a key role in myocardial injury.[26] On the other hand, other factors, such as hypoxemia induced by severe pneumonia,[27,28] psychological stress and anxiety in the patients can also cause hypoxic-ischemic heart damage.[29-33] Thus, the pathogenesis of cardiac injury in patients with SARS-CoV-2 infection remains to be further explored.

This study has several limitations. First, owing to limited medical resources, only 28 severe patients with COVID-19 pneumonia were included during this period. Second, this study was conducted at a single-center hospital with limited sample size and most patients came from Hubei province. There may also be a selection bias when analyze factors for the severity of disease. Third, this is a retrospective study and the data permit a preliminary assessment of the clinical course and outcomes of patients with COVID-19. A larger, multicenter cohort study of patients with COVID-19 would help to further clarify the clinical characteristics and risk factors of the disease.

5 | CONCLUSIONS

In this study, we found that the duration of fever, C-reactive protein and PO_2 < 80 mm Hg on admission were associated with the severity of COVID-19 in the early stage, which may be beneficial for clinicians to evaluate the disease severity and provide a reference for evaluating therapeutic effect. SARS-CoV-2 viral RNA could be long-term presence in respiratory tract and fecal sample and there is no correlation between the viral shedding and the severity of COVID-19.

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CONFLICT OF INTERESTS

The authors declare that there are no conflict of interests.

AUTHOR CONTRIBUTIONS

XZ, JC, LD, GC, ZF, and DY collected the epidemiological and clinical data. XZ, JC, and ZH analyzed and summarized all data. XZ drafted the manuscript. ZH and JX revised the final manuscript.

DATA AVAILABILITY STATEMENT

The data that supports the findings of this study are available in this published article and its the supplementary material.

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section.

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