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Severe COVID-19 patients with liver injury: a seven-case series

X. LI[1-3], Z.-C. ZHANG[2,3], P.-L. ZHANG[4]

[1]Department of Neurology, The Third Central Clinical College of Tianjin Medical University, Tianjin, China
[2]Department of Neurology, Tianjin Third Central Hospital; The Third Central Hospital of Tianjin, Tianjin, China
[3]Tianjin Key Laboratory of Extracorporeal Life Support for Critical Diseases, Tianjin, China
[4]Department of Internal Neurology, Tianjin Huanhu Hospital, Tianjin, China

Abstract. **We present the case details of seven patients diagnosed with severe novel coronavirus disease 2019 (2019-nCoV, hereafter COVID-19) with hepatic injury. Most of these patients were elderly and had hypertension, diabetes mellitus, coronary heart disease, and other underlying health conditions prior to admission for COVID-19. Liver injury occurred in all seven cases during the course of the disease. Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels initially increased (1.2-times to 2.0-times the normal value, respectively) in the second week. The liver function recovered in all patients within one week of conventional liver protection treatment. Elevated serum transaminase levels in these patients were due to the COVID-19 infection but could also be related to systemic immune response caused by cytokine storm syndrome (CSS) and hepatocyte damage caused by ischemia and hypoxia. COVID-19 is highly infectious and mainly affects the lungs. In some cases, especially in patients with severe disease type, COVID-19 may also cause liver injury. The liver function of patients with severe COVID-19 should be very carefully monitored, especially if the patients are elderly and have underlying comorbidities.**

Key Words:
2019 Novel coronavirus (2019-nCoV), Coronavirus disease 2019 (COVID-19), Severe disease, Liver injury.

Introduction

The outbreak of novel coronavirus disease caused by the 2019 novel corona virus (2019-nCoV) is a serious infectious disease that has led to a global pandemic. The World Health Organization (WHO) named the disease caused by this novel pathogens "COVID-19" on February 11, 2020[1]. In addition to respiratory symptoms, such as fever, dry cough, and dyspnea, COVID-19 patients also showed different degrees of liver injury. With the rapidly increasing number of cases, COVID-19 with concomitant liver injury has received more attention from clinicians. Herein, we report the case details of seven patients with severe COVID-19 infection with liver injury.

This study was registered and conducted in Tianjin Third Central Hospital in February 2020 and was approved by the Hospital's Ethics Committee. The Ethics Committee also approved related screening, treatment, and data collection of these patients. All subjects signed the written informed consent form. The study protocol was in accordance with the tenets of the Declaration of Helsinki.

Case Reports

We included seven patients (3 male and 4 female; mean age, 61.1±13.6 years) with severe convalescent COVID-19, who were treated in Tianjin Jinnan Hospital from February 14, 2020 to February 26, 2020. Tianjin Jinnan Hospital is the fourth designated hospital in Tianjin, specifically for the treatment of COVID-19 cases.

All seven patients had a close contact history with confirmed or suspected COVID-19 patients. COVID-19 was confirmed in all patients by positive results of Reverse Real Time-Polymerase Chain Reaction (RT-PCR) from pharyngeal swab specimens.

Corresponding Author: Li-Xia, MD; e-mail: doctorlixia@tmu.edu.cn

The first presenting symptom in all seven patients was fever. While five out of seven patients presented with progressively worsening dry cough and dyspnea in the short term, the remaining two patients did not have cough initially, but their disease rapidly worsened from shortness of breath to dyspnea directly. All seven patients suffered from dyspnea during the course of the disease and two patients had pharyngalgia.

Most of these patients were elderly; two had a history of coronary heart disease, including one with a history of paroxysmal atrial fibrillation, two had diabetes mellitus, and four had hypertensions.

None of the seven patients had any underlying liver disease, and their hepatitis markers were negative. The results of their liver function tests were initially normal, but the leukocyte and peripheral blood lymphocyte counts were decreased. Chest computed tomography (CT) showed multiple ground glass opacities and infiltration shadows in both lungs, with evident extrapulmonary zones (Figure 1). The D-dimer value was elevated in two patients (Table I).

According to COVID-19's Prevention and Control Plan (Fourth Edition)[2], severe COVID-19 was defined when the patients met any of the following criteria: (1) respiratory frequency ≥ 30 times/min; (2) blood oxygen saturation ≤ 93% at rest; (3) partial pressure of arterial oxygen (PaO_2) to fraction of inspired oxygen ratio (FiO_2) ≤ 300 mmHg (1 mmHg = 0.133kPa); (4) and lung infiltrates > 50% within 24 hours to 48 hours. After expert consultation, the clinical classification of the seven patients was identified as severe COVID-19.

All seven patients were treated with lopinavir/ritonavir 500 mg BID, Arbidol 0.2 g TID, and interferon-alpha aerosol antiviral therapy after admission. Three patients received short-term glucocorticoid therapy (3-5 days) as appropriate to alleviate the inflammatory response. Liver injury was observed in all seven cases during the course of the disease. Serum alanine aminotransferase (ALT) and aspartate aminotransferase (AST) levels were initially increased (1.2-times to 2.0-times the normal value, respectively) in the second week; ALT levels ranged between 60 U/L and 102 U/L (normal range: 4 U/L-44 U/L), and AST levels ranged between 40 U/L and 65 U/L (normal range: 8U/L-38U/L). Furthermore, five patients had hypoalbuminemia of 30.2 g/L-34.7 g/L (normal range: 35 g/L-50 g/L). All seven patients with liver injury were transferred from Tianjin Haihe Hospital to Tianjin Jinnan hospital for further consolidated treatment.

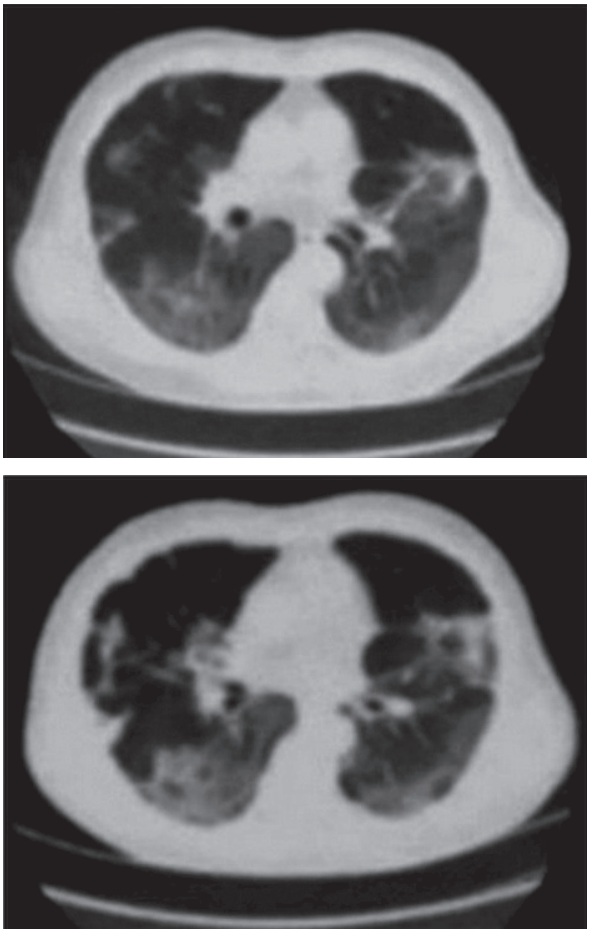

Figure 1. Chest computed tomographic (CT) manifestations of patient 3. Multiple patch-like shadows and ground-glass opacity in both lungs.

Furthermore, all seven patients were re-examined twice, 24-h apart, before being discharged; and the results of reverse real-time PCR assay from pharyngeal swabs were negative. The mean duration of hospitalization in Tianjin Haihe Hospital was 19±4.6 days (range: 11-24 days).

During hospitalization in Tianjin Jinnan hospital, two out of seven patients had diarrhea, i.e., 3-4 episodes per day of loose stools, and one episode of constipation. All seven patients received nutritional support treatment and were given oral polyene phosphatidylcholine capsules 456 mg (TID) to protect the liver, two out of seven patients additionally received oral bicyclol tablets 50 mg (TID) for hepatoprotection. After a week of treatment, all patients' liver function returned to baseline value. The patients recovered and were discharged.

Table I. Clinical data of the seven COVID-19 patients.

Variables	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5	Patient 6	Patient 7
Age (years)	68	47	55	77	46	56	79
Sex	Female	Male	Male	Female	Female	Female	Male
Exposure history	All close contact with COVID-19 patients						
Anamnesis							
Coronary heart disease				Yes		Yes	
Diabetes mellitus		Yes					Yes
Hypertension	Yes	Yes		Yes			Yes
Disease duration (d)	23	22	15	19	11	19	24
Clinical types	Severe	Severe	Severe	Severe	Severe	Severe	Severe
Symptoms							
Fever	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Cough	Yes		Yes	Yes	Yes		Yes
Pharyngalgia		Yes			Yes		
Nausea/vomiting					Yes		
Diarrhea	Yes		Yes				
Constipation					Yes		
Dyspnea	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Albumin (g/L)	34.7	35.1	33.6	34.6	32.3	34.2	30.2
Alanine aminotransferase (U/L)	66	89	84	60	102	77	70
Aspartate aminotransferase (U/L)	45	52	56	40	65	46	33
Leukocyte count ($\times 10^9$/L)	2.3	2.96	2.39	3.67	5.59	3.67	2.92
Lymphocyte count ($\times 10^9$/L)	0.43	0.78	0.49	0.83	1.6	0.83	0.63
Neutrophils ($\times 10^9$/L)	1.78	2.05	1.69	2.48	3.23	2.32	0.216
Platelets ($\times 10^9$/L)	135	137	131	226	280	224	83
Hemoglobin (g/L)	128	158	136	102	131	134	131
D-dimer (mg/L)	0.2	0.3	1.7	0.4	0.3	0.4	2
Chest CT	bilateral pneumonia multiple ground glass shadow, infiltration shadow, with the lung outer zone evident						
Treatment							
Lopinavir/ritonavir	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Arbidol	Yes	Yes	Yes	Yes	Yes	Yes	Yes
α-interferon	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Glucocorticoid		Hydrocortisone sodium succinate	Methyl prednisolone		Methyl prednisolone		Dexa-methasone
Bicyclol tablets	Yes				Yes		
Polyene phosphatidyl -choline capsules	Yes	Yes	Yes	Yes	Yes	Yes	Yes

Discussion

Patients with COVID-19 are prone to concomitant liver injury during treatment. Chen et al[3] reported on this issue in 99 patients with the novel COVID-19 infection. They mentioned that the liver function indicators, such as serum AST, ALT, total bilirubin (TBil), and lactic dehydrogenase (LDH) inpatients admitted to intensive care were significantly higher than those not in intensive care.

All seven patients with severe COVID-19 showed liver injury, which were consistent with previous studies[3-6]. At the same time, this study also showed that COVID-19 pneumonia with liver injury as the first manifestation is relatively infrequent, whereas secondary liver injury is more common. Furthermore, the liver function of these seven patients was normal at admission, and liver injury occurred after a certain period of hospitalization. COVID-19 first affected the lungs and subsequently, the liver.

Current clinical data show that the proportion of critically ill patients with COVID-19 with hypertension and diabetes is high[3-6]. Four of our seven patients had hypertension, two had diabetes, and one had coronary heart disease, which is consistent with previous reports. Although all seven patients had hepatic impairment, their effect was mild and the prognosis improved after timely treatment.

Currently, studies on the mechanisms of COVID-19-related liver injury are limited. Liver injury in COVID-19 may be directly caused by the virus itself; it could also be related to immune damage, inflammation-induced systemic immune response, and/or hepatocyte ischemia or hypoxia.

2019-nCoV Infection of Hepatocytes Leads to Liver Injury

Human angiotensin-converting enzyme 2 (ACE2) is the receptor for the 2019-nCoV and facilitates its cell entry. As alveolar type 2 cells highly express ACE2, the lung is the primary target organ of 2019-nCoV. Studies have found that the biliary endothelial cells of liver tissue show2019-nCoV receptor-ACE2 expression; therefore, it is speculated that 2019-nCoV can infect biliary endothelial cells, cause inflammatory injury to the liver, and eventually lead to abnormal elevation of liver enzymes[7].

Previous research has reported that the upregulation of ACE2 expression in liver tissue, caused by compensatory proliferation of hepatocytes was derived from bile duct epithelial cells, and may also be responsible for liver tissue injury caused by 2019-nCoV infection[8].

Liver biopsy specimens from patients with COVID-19 showed moderate microvascular steatosis, mild lobular and portal activity, indicating that the injury could have been caused by either the 2019-nCoV infection directly or drug-induced liver injury[9].

The COVID-19's Prevention and Control Plan (7th edition) initialized by The General Office of The State Health Commission of China has reported the following pathological changes in the liver and gallbladder: increased size, dark-red appearance, hepatocyte degeneration, focal necrosis with neutrophil infiltration, hepatic sinusoidal congestion, lymphocyte and monocyte infiltration in the portal area, microthrombosis, and gallbladder filling[10].

All seven patients had severe COVID-19 illness and mainly showed mild elevation of ALT and AST, although the biochemical markers of bile duct injury, i.e., ALP and GGT, were not significantly elevated. Jaundice is rare though hypoalbuminemia is more frequent. Therefore, abnormal liver function indicators in COVID-19 patients are more likely caused by the toxic effects of 2019-nCoV, and may also be related to systemic inflammatory and hepatocyte ischemia or hypoxia.

In addition, it should not be forgotten that COVID-19 is high-risk infectious disease, which may also attack other organs, such as the heart, kidney, and brain. Huang et al[4] reported that 5 of 41 patients with COVID-19 had acute cardiac injury. Chen et al[3] in their study on 99 patients with the novel COVID-19 infection reported that most patients had abnormal myocardial zymogram, which showed an elevation of creatine kinase and LDH in13 (13%) and 75 (76%) patients, respectively; one of these patients also showed abnormally elevated creatine kinase (6,280 U/L) and LDH (20, 740 U/L). Seven (7%) patients had different degrees of renal dysfunction, with elevated blood urea nitrogen or serum creatinine[3]. In our patients, elevated serum ALT was more pronounced which predominantly suggested liver injury.

Immune Damage and Inflammation-Induced Systemic Immune Response

Liver injury in COVID-19 patients may be related to the systemic immune response and immune injury caused by the viral infection. In our

clinical setting, we found that liver injury often occurred in patients around the second week of active infection, which indicated that it was most likely related to the patient's immune status. Liver injury may be related to the inflammatory storm caused by virus-induced injury. Cytokine storm syndrome (CSS) is a phenomenon that is dysregulated in the immune system owing to the dramatic increase of proinflammatory cytokine levels, after the body is stimulated by microorganisms or drugs[11]

In the process of resisting viral infection, specific immune and non-specific immune responses interact with each other and cooperate closely to yield immune protection. However, specific immune response is the key factor for complete virus clearance[12]. If the human body fails to produce a strong-enough specific immune response to effectively kill the virus after it invades the host cells, the immune system will continue to strengthen the non-specific inflammatory response to inefficiently kill the virus. However, this phenomenon cannot help eliminating 2019-nCoV; on the contrary, it aggravates the infection and leads to tissue ischemia, hypoxia, and even necrosis. Eventually, it can lead to uncontrolled non-specific inflammatory response and trigger a CSS.

The occurrence of CSS will not only lead to lung injury but also cause injury to the liver, kidney, myocardium, and other organs and tissues. Elderly patients take a long time to form a specific immune response owing to reduced immunity; therefore, they are very likely to develop CSS, which may further worsen the seriousness of the disease.

Our case series shows that the early application of glucocorticoid alleviates severe immune response in some patients, which, along with hepatoprotective drugs, helps the liver function of patients to return to baseline. However, our sample size is quite small, and more in-depth studies are needed to further explore the causes of liver injury in patients with COVID-19 infection.

Hepatic Ischemia, Hypoxia, and Hypoperfusion Also Aggravate Liver Injury

Severely and critically ill COVID-19 patients suffer from extensive pulmonary inflammation, microcirculation disturbance, and decreased oxygenation capacity. As a result, tissue hypoxia and accumulation of lactic acid affects the acid-base balance and aggravates internal environment disorders.

Previous studies have found that hepatocyte death and inflammatory cell infiltration, both caused by ischemia and hypoxia, can be found in liver transplantation specimens, both *in vivo* and *in vitro* models of hepatic ischemia and hypoxia[13]. COVID-19 patients have different degrees of hypoxemia, wherein > 40% of them need oxygen therapy[6]. All seven of our patients had hypoxemia and required oxygen therapy.

Furthermore, cardiac preload increases in critically ill COVID-19 patients because of pulmonary inflammation and consolidation. If patients have concomitant cardiovascular diseases, are older, and have other comorbid conditions, the poor compensatory ability of heart function will lead to congestion of hepatic sinuses in portal hypertension. Thus, it can aggravate ischemia and hypoxia of hepatocytes, affect the discharge of toxic metabolites, and further aggravate the damage to liver function.

As all seven patients in this report were aged between 46 and 79 years and suffered from underlying health conditions, they were prone to microcirculation disorders, and portal hypertension caused liver ischemia, hypoxia, and perfusion insufficiency and injury.

Therefore, ischemia and hypoxia may be one of the main mechanisms of liver injury in patients with severe COVID-19 infection.

Conclusions

COVID-19 is highly infectious and mainly affects the lungs. All seven patients in our report had severe type COVID-19 and showed complicated liver injury.

Our findings suggest that we should pay close attention to complications related to liver injury during the treatment of COVID-19 patients.

It is important to observe the liver function of COVID-19 patients, especially in elderly patients. Severely ill patients with hypertension, diabetes, coronary heart disease, and other underlying health complications should be monitored very closely for liver function, and the occurrence of liver injury, and timely hepatoprotective therapy should be provided.

Conflict of Interests

The authors declare that they have no conflict of interests.

References

1) World Health Organization (WHO). Coronavirus disease (COVID-2019) situation reports 2020. Available at https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/naming-the-coronavirus-disease-(covid-2019)-and -the-virus-that-causes-it. Accessed.
2) Circular of The General Office of The State Health Commission on Issuing COVID-19's Prevention And Control Plan (Fourth Edition). National Health Commission of the People's Republic of China 2020. Available at http://www.gov.cn/zhengce/zhengceku/2020-01/28/5472673/files/0f96c10cc09d4d36a6f9a9f0b42d972b.pdf [Chinese].
3) Chen N, Zhou M, Dong X, Qu J, Gong F, Han Y, Qiu Y, Wang J, Liu Y, Wei Y, Xia J, Yu T, Zhang X, Zhang L. Epidemiological and clinical characteristics of 99 cases of 2019 novel coronavirus pneumonia in Wuhan, China: a descriptive study. Lancet 2020; 395: 507-513.
4) Huang C, Wang Y, Li X, Ren L, Zhao J, Hu Y, Zhang L, Fan G, Xu J, Gu X, Cheng Z, Yu T, Xia J, Wei Y, Wu W, Xie X, Yin W, Li H, Liu M, Xiao Y, Gao H, Guo L, Xie J, Wang G, Jiang R, Gao Z, Jin Q, Wang J, Cao B. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. Lancet 2020; 395: 497-506.
5) Wang D, Hu B, Hu C, Zhu F, Liu X, Zhang J, Wang B, Xiang H, Cheng Z, Xiong Y, Zhao Y, Li Y, Wang X, Peng Z. Clinical characteristics of 138 hospitalized patients with 2019 novel coronavirus-infected pneumonia in Wuhan, China. JAMA 2020; 323: 1061-1069.
6) Guan WJ, Ni ZY, Hu Y, Liang WH, Ou CQ, He JX, Liu L, Shan H, Lei CL, Hui DSC, Du B, Li LJ, Zeng G, Yuen KY, Chen RC, Tang CL, Wang T, Chen PY, Xiang J, Li SY, Wang JL, Liang ZJ, Peng YX, Wei L,Liu Y, Hu YH, Peng P, Wang JM, Liu JY, Chen Z, Li G, Zheng ZJ, Qiu SQ, Luo J, Ye CJ, Zhu SY, Zhong NS; China Medical Treatment Expert Group for Covid-19. Clinical characteristics of coronavirus disease 2019 in China. N Eng J Med 2020;382:1708-1720.
7) Chai XQ, Hu LF, Zhang Y, Han WY, Lu Z, Ke AW, Zhou J, Shi GM, Fang N, Fan J, Cai JB, Fan J, Lan F. Specific ACE2 expression in cholangiocytes may cause liver damage after 2019-nCoV infection. BioRxiv 2020. [Epub ahead of print] DOI: 10.1101/2020.02.03.931766V1 .
8) Guan GW, Gao L, Wang JW, Wen XJ, Mao TH, Peng SW, Zhang T, Chen XM, Lu FM. Exploring the mechanism of liver enzyme abnormalities in patients with novel coronavirus-infected pneumonia. Chin J Hepatol 2020; 28: 100-106.
9) Xu Z, Shi L, Wang Y, Zhang J, Huang L, Zhang C, Liu S, Zhao P, Liu H, Zhu L, Tai Y, Bai C, Gao T, Song J, Xia P, Dong J, Zhao J, Wang FS. Pathological findings of COVID-19 associated with acute respiratory distress syndrome. Lancet Respir Med 2020; 8: 420-422.
10) Circular of the General Office of The State Health Commission on Issuing COVID-19's Prevention And Control Plan (Seventh Edition). National Health Commission of the People's Republic of China 2020. Available at http://www.nhc.gov.cn/yzygj/s7653p/202003/46c9294a7dfe4cef80dc7f5912eb1989/files/ce3e6945832a438eaae415350a8ce964.pdf [Chinese].
11) Shimabukuro-Vornhagen A, Gödel P, Subklewe M, Stemmler HJ, Schlösser HA, Schlaak M, Kochanek M, Böll B, von Bergwelt-Baildon MS. Cytokine release syndrome. J Immunother Cancer 2018; 6: 56.
12) Akiko Iwasaki, Ruslan Medzhitov. Regulation of adaptive immunity by the innate immune system. Science 2010; 327: 291-295.
13) Yang L, Wang WJ, Wang XZ, Zhao JF, Xiao L, Gui WF, Fan HQ, Xia J, Li ZL, Yan JG, Afnan Alasbahi, Zhu QJ, and Hou XH. Creg in Hepatocytes ameliorates liver ischemia/reperfusion injury in a TAK1-Dependent manner in mice. Hepatology 2019; 69: 294-313.