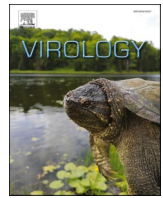




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Effect of SARS-CoV-2 coinfection was not apparent on the dynamics of chronic hepatitis B infection

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ABSTRACT

In patients coinfecting with SARS-CoV-2 and HBV, liver injury was common. However, the interactions between SARS-CoV-2 and HBV coinfection remained unknown. Sixty-seven COVID-19 patients from the previous cohort were enrolled and classified into 2 groups (7 with HBsAg+ and 60 with HBsAg-). The association of HBV- and SARS-CoV-2-related markers were analyzed. During the acute course of SARS-CoV-2 infection, markers of HBV replication did not extensively fluctuate during SARS-CoV-2 infection. Coinfection with HBV did not extend the viral shedding cycle or incubation periods of SARS-CoV-2. Effects of SARS-CoV-2 on the dynamics of chronic HBV infection seemed not apparent. SARS-CoV-2 infection would not be the source of HBV reactivation in these individuals.

1. Introduction

Elevated aminotransferases and bilirubin were common in COVID-19 patients (Hao et al., 2020; Fu et al., 2020) and typical characteristics of patients with chronic hepatitis B (CHB). In patients coinfecting with SARS-CoV-2 and HBV, Zou et al. reported a proportion of 27.62% with elevated liver function tests (Zou et al., 2020), which were similar to SARS-CoV-2 infection alone (Huang et al., 2020). However, these reports failed to focus on the direct interactions between SARS-CoV-2 and HBV coinfection. Especially, to what degree the viral kinetics, immune response, or natural history of the two viruses and their impact on disease progression would be influenced remained unknown.

2. Methods

Sixty-seven COVID-19 patients from our previous prospective cohort

(Tan et al., 2020) were enrolled in this study. According to the status of HBsAg, they were classified as HBsAg + group (n = 7) and HBsAg- group (n = 60). COVID-19 was confirmed by real-time PCR assay. Clinical and laboratory data were collected on day 1, 4, 7, 14, 18, 21, 28, if available, after admission and at discharge. All 7 HBsAg + COVID-19 patients had reported chronic HBV infection (anti-virus treatment-free) for more than 20 years with normal liver function tests always, except that patient 37 developed HBV-related decompensation liver cirrhosis 3 years ago.

3. Results

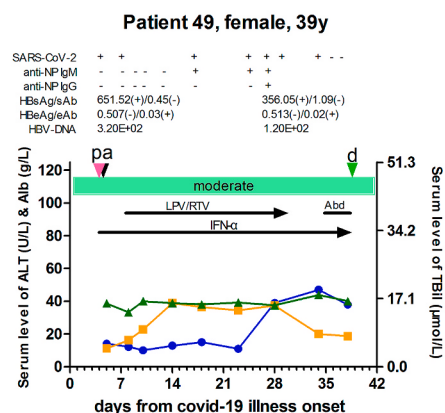
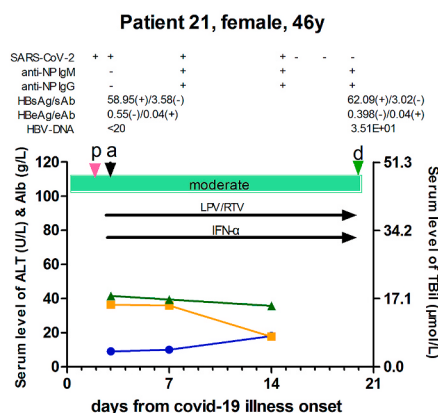
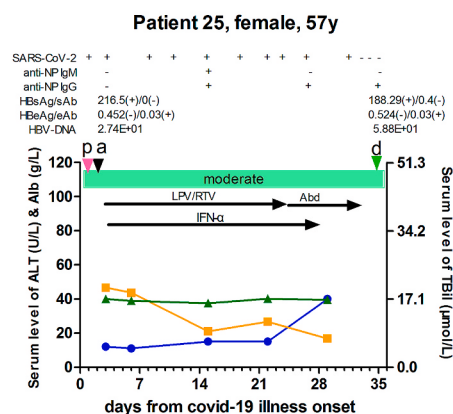
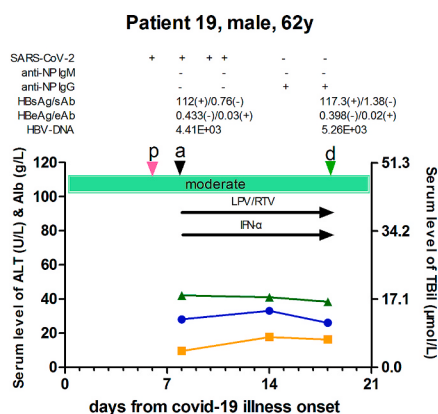
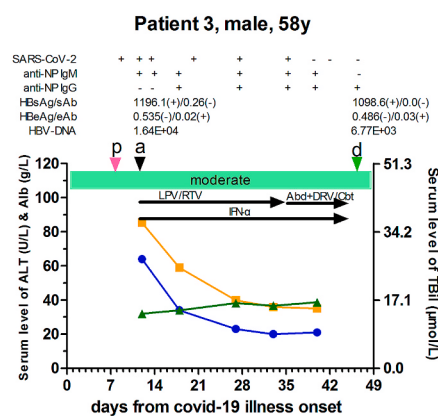
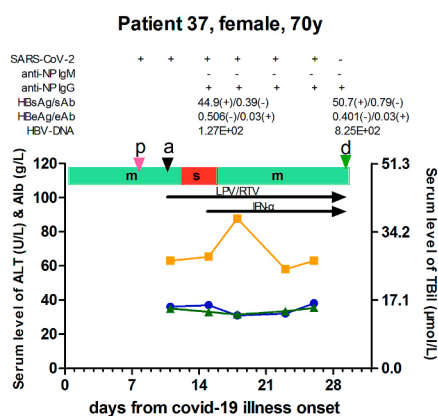
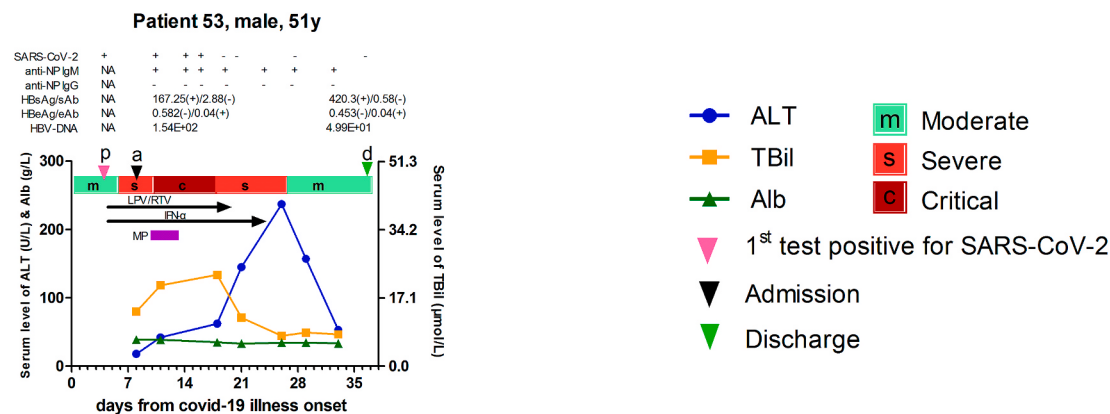
The basic characteristics were described in Table S1. Briefly, there were 3 males and 4 females enrolled, including 1 critical case, 3 severe cases, and 3 mild/moderate cases, with the median age of 57 yrs (39–70). In the HBsAg + group, 6 of them were HBsAg-negative chronic infection based on EASL recommendation (6) and 1 of them were HBV-

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Fig. 1. Sequential SARS-CoV-2- and HBV-related parameters along with serum hepatic enzyme markers for 7 chronic hepatitis B patients after acute new coronavirus infectious. The color bar shows the severity of pneumonia from moderate, severe, to critical. Black arrows indicate the administration of antiviral drugs against SARS-CoV-2. HBV DNA was determined by COBAS Amplicor monitor test (Roche Molecular Systems, Branchburg, NJ), meanwhile, the HBsAg, anti-HBs, HBeAg, and anti-HBe were detected quantitatively by chemiluminescent immunoassay (Abbott Laboratory, Chicago, IL). ALT, alanine transaminase; TBil, total bilirubin; Alb, Albumin; NP, nucleocapsid protein; LPV/RTV, lopinavir/ritonavir; IFN- α 1b, interferon- α 1b; MP, methylprednisolone; Abd, arbidol; DRV/Cbt, darunavir/cobicistat.

Table 1

The COVID-19 progression, the intensity of anti-NP response, and liver injury after acute SARS-CoV-2 infection in patients with or without chronic hepatitis B.

	All patients (n = 67)	COVID-19 patients with HBsAg positive (n = 7)	COVID-19 patients without HBV (n = 60)	p-value
HBsAg positive	7 (10.4)	7 (100.0)	0	–
HBeAg positive	0	0	0	–
Comorbidity besides Hepatitis B	21/67 (31.3)	2/7 (28.6)	19/60 (31.7)	>0.999
Pulmonary disease	3 (4.5)	1 (14.3)	2 (3.3)	
†				
Diabetes and/or Hypertension	15 (22.3)	1 (14.3)	14 (23.3)	
Hyperlipemia and/or CHD	3 (4.5)	0	3 (5.0)	
Liver injury at admission #	5/67 (7.5)	0	5/60 (8.3)	>0.999
Liver injury admission & during hospitalization	19/67 (28.4)	3/7 (42.9)	16/60 (26.7)	0.395
Liver injury type #				0.432
Hepatocellular	3/67 (4.5)	1/7 (14.3)	2/60 (3.3)	
Ductular	11/67 (16.4)	2/7 (28.6)	9/60 (15.0)	
Mix	5/67 (7.5)	0	5/60 (8.3)	
COVID-19 progression after admission	14/67 (20.9)	2/7 (28.6)	12/60 (20.0)	0.63
COVID-19 stable after admission	53/67 (79.1)	5/7 (71.4)	48/60 (80.0)	0.63
Shedding time of SARS-CoV-2, days*	25.0 $\pm$ 9.4	27.1 $\pm$ 9.0	24.7 $\pm$ 9.5	0.52
anti-NP-IgM development	28/58 (48.3)	4/7 (57.1)	24/51 (47.1)	0.701
weak-response	10/58 (17.2)	1/7 (14.3)	9/51 (17.6)	>0.999
strong-response	18/58 (31.0)	3/7 (42.9)	15/51 (29.4)	0.665
Days of anti-NP-IgM first detectable §	12.3 $\pm$ 4.4	12.5 $\pm$ 3.7	12.2 $\pm$ 4.6	0.909
anti-NP-IgG development	45/54 (83.3)	6/7 (85.7)	39/47 (83.0)	>0.999
weak-response	33/54 (61.1)	4/7 (57.1)	29/47 (61.7)	>0.999
strong-response	12/54 (22.2)	2/7 (28.6)	10/47 (21.3)	0.645
Days of anti-NP-IgG first detectable §	14.3 $\pm$ 4.9	15.8 $\pm$ 6.2	14.0 $\pm$ 4.7	0.405
Duration of Hospitalization, days	21.0 $\pm$ 9.1	25.0 $\pm$ 10.7	20.6 $\pm$ 8.9	0.228
COVID-19 course, days	27.6 $\pm$ 9.2	32.0 $\pm$ 10.4	27.0 $\pm$ 9.0	0.178
Discharge	67/67 (100.0)	7/7 (100.0)	60/60 (100.0)	>0.999

† Includes one patient with bronchiectasis and chronic bronchitis, one patient with pulmonary tuberculosis, and one patient with asthma. CHD, coronary heart disease. # In this study, we defined ALT and/or AST more than 3 times of the upper limit units (ULN), GGT, and/or TBIL more than 2 $\times$ ULN as liver injury. ALT and/or AST over 3 $\times$ ULN were classified as hepatocellular type, GGT and/or TBIL over 2 $\times$ ULN as ductular type, while combined with ALT and/or AST over 3 $\times$ ULN and GGT and/or TBIL over 2 $\times$ ULN as mix type. * Virus shedding in any type of sample, including nasopharyngeal swab, sputum, and stool. § Days from symptom onset. Data are mean $\pm$ standard deviation or n/N (%), where N is the total number of patients with available data.

related cirrhosis There were no significant differences in demographic and epidemiological characteristics between the two groups, except median CRP levels. But there was no difference in the rate of patients with CRP elevation. Besides, there were no significant differences in the clinical severities, duration of hospitalization, incubation periods, and treatment response, either. At admission, levels of alanine transaminase (ALT) in 17 (25.4%) cases were over 50IU/L, while 28.4% had a TBil level over 17.1 μ mol/L. However, no significant difference was detected between patients with HBsAg positive and negative (Table S1). At discharge, levels of ALT and TBil decreased to 32.3 $\pm$ 21.9U/L and 13.3 $\pm$ 6.7 μ mol/L, but there were still about 15% of patients with abnormal liver indices. Likewise, there was no difference in clinical indices between the two groups at discharge (Table S2). Extreme values (peak or trough values) of each index were also compared, but no significant differences were detected (Table S3).

HBV-related markers were measured at both admission and discharge, but there were no significant differences in all markers, including HBsAg, HBsAb, HBeAg, HBeAb, HBcAb, and HBV-DNA (Table S4) at the beginning of and after recovery from COVID-19 for chronic hepatitis B (CHB) cases. Apart from that, sequential serum levels of HBV- and SARS-CoV-2-related parameters along with hepatic enzyme markers were analyzed in each of the 7 patients (Fig. 1). Firstly, there were no obvious changes in the serum level of HBsAg/Ab, HBeAg/Ab, or HBV-DNA viral load been observed during the course of acute SARS-CoV-2 infection and/or the anti-nucleocapsid protein (NP) antibody development in these 7 patients. Secondly, serum levels of ALT and TBil in CHB patients did not change before and after hospitalization either. Particularly, patient 53 presented a liver injury since day 18, which was considered just as the side-effect of lopinavir/ritonavir (LPV/RTV), and ALT decreased to normal after LPV/RTV withdrawal. Patient 37 had an abnormal serum TBil level, which we thought was attributed to HBV-related decompensated liver cirrhosis for more than 3 years. Thirdly, our results showed that the administration of antiviral drugs, like LPV/RTV, arbidol, and interferon- α 1b, which were prescribed by clinicians attempting to inhibit SARS-CoV-2 in the initial stage, seemed to have no affection on the replication of HBV. The other way round, the pre-existing HBV in the host body seemed to have no effect on COVID-19 progression, anti-NP antibody development, the intensity of anti-NP response, and even the liver injury after acute SARS-CoV-2 infection (Table 1). All 67 patients were discharged from the hospital with recovery.

4. Discussion

In this study, we reported the sequential levels of HBV-related markers during the acute infection of SARS-CoV-2. Previous studies on patients coinfecting with SARS-CoV-2 and HBV have shown that the existence of coinfection could not aggravate the liver injury or extend the duration of hospitalization compared with HBV infection alone (Chen et al., 2020; Liu et al., 2020). However, these studies failed to depict levels of all HBV-related markers during the infection and clearance of SARS-CoV-2 infection. In this study, although just 7 cases were enrolled, we described the intact dynamics of these markers, indicating that SARS-CoV-2 infection had a limited influence on HBV-DNA replication or the natural history of HBV infection. Firstly, the quantitative levels of HBsAg/Ab, HBeAg/Ab, and HBV-DNA did not extensively fluctuate during the infection or clearance of SARS-CoV-2, suggesting that SARS-CoV-2 had no impact on HBV kinetics. Secondly, the coinfection of SARS-CoV-2 did not trigger the reactivation or the

seroconversion of chronic hepatitis B, which is also reported in Rodríguez-Tajes S et al.'s study (Rodríguez-Tajes Sergio et al., 2020). Consequently, SARS-CoV-2 infection would not be the source of HBV reactivation in these individuals. On the other hand, the existence of HBV did not influence SARS-CoV-2, either. Virologically, coinfection with HBV did not extend the viral shedding cycle or incubation periods of SARS-CoV-2 infection. Clinically, the coinfection of HBV did not increase the severity of diseases or duration of hospitalization. At discharge, no significant differences could be detected between the two groups in disease recovery, either. Besides, it has been reported that HBV antiviral therapy had a limited effect on COVID-19 incidence and outcomes (Sabela et al., 2020). From both sides, we discussed the interactions between HBV and SARS-CoV-2 infection.

Apparently, hepatic impairment could be detected in some COVID-19 patients. But we thought it was more likely to be the consequence of illness severity, rather than the direct cytotoxic effect of SARS-CoV-2 on the liver. Consistent with our conclusions, previous larger sample studies also found that there was no significant difference in liver enzymes between coinfection and HBV infection alone (7,8). The elevations of the liver enzyme in COVID-19 patients could also happen in many respiratory viruses infection, which might be related to immune interactions involving systematic cytokine storms (Adams and Hubscher, 2006). Apart from that, sarcolysis during acute viral infection (Wang and Medzhitov, 2019) and drug-induced liver injury (Marta et al., 2020) could also explain the elevation of liver enzymes.

However, it's undeniable that this is a preliminary study enrolling just 7 patients coinfecting with SARS-CoV-2 and HBV in a single center. Although detailed patients' information was collected, retrospective design restricted the further analysis. For example, only 1 out of 7 patients had a history of liver cirrhosis, and COVID-19 is reported to be associated with liver function deterioration and elevated mortality in patients with cirrhosis and COVID-19 (Massimo et al., 2020). As a result, we could hardly know whether coinfection had a different story in patients with liver cirrhosis in our study. Consequently, further large-scale studies should be conducted to reveal the exact interactions between SARS-CoV-2 and HBV coinfection.

Above all, our study suggested that the effects of SARS-CoV-2 on the dynamics of chronic HBV infection seemed not apparent. SARS-CoV-2 infection would not be the source of HBV reactivation in these individuals. More attention should be paid to viral control, immune modulation, and reasonable medication in the therapy of patients coinfecting with HBV and SARS-CoV-2.

Specific author contributions

GD, WT, YC; Acquisition of data: ST, YD, YL, JZ, ZT, XH, XX, YZ, YG, WT, YC; Statistical analysis: WT; Interpretation of data: RY, WT, GD; Drafting and critical revision of manuscript: RY, WT, GD. All authors checked and approved the final draft of the letter.

Guarantor of the article

Wenting Tan, Ph.D., Yaokai Chen, Ph.D., and Guohong Deng, Ph.D.

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Declaration of competing interest

Potential competing interest: None to report.

Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.virol.2020.11.012>.

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