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Declined serum high density lipoprotein cholesterol is associated with the severity of COVID-19 infection

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ABSTRACT

Background: COVID-19 infection is epidemic worldwide. We describe the serum lipid profile of the patients with COVID-19 infection.
Methods: In this retrospective study, we collected the first clinical laboratory data of 114 patients on admission, and 80 healthy controls. Meanwhile, we monitored the serum lipid profile, COVID-19 nucleic acid and chest CT scan of a severe patient from the early stage of infection to the recovery period for a total of 80 days.
Results: Compared with the healthy controls, the patients had sharply decreased concentrations of total cholesterol, HDL-cholesterol and LDL-cholesterol ($P < 0.001$). Among the patients, HDL-cholesterol concentration in severe groups was significantly lower than the common groups [1.01 (0.88–1.20) *vs* 1.21 (1.02–1.48) mmol/l, $P < 0.001$]. The lipid profile of a severe patient showed that serum cholesterol concentration significantly decreased in the early stage and returned to be normal in the recovery period. Moreover, the change of HDL-cholesterol in this patient was consistent with the results of nucleic acid tests and chest CT scans. In correlation analysis, HDL-cholesterol concentration was negatively correlated with C-reactive protein (CRP, $r = -0.396$, $P < 0.001$) and positively correlated with lymphocytes ($r = 0.336$, $P < 0.001$). The area under curve (AUC) in receiver operating characteristic (ROC) of HDL-cholesterol was 0.732 ($P < 0.001$), and, the adjusted odd ratio (OR) of HDL-cholesterol was 0.023 (95% CI 0.002–0.227).
Conclusions: Decreased serum HDL-cholesterol is associated with the severity of COVID-19 infection.

1. Introduction

Coronaviruses are a large family of viruses known to cause clinical symptoms ranging from the common colds to sever lung infections, such as Severe Acute Respiratory Syndrome (SARS) and Middle East Respiratory Syndrome (MERS), which caused by SARS-CoV and MERS-CoV respectively. COVID-19 is a novel strain of coronavirus, and has been identified as the causal pathogen of an ongoing worldwide epidemic in 2020. COVID-19 are enveloped, non-segmented positive-strand RNA viruses, and cause Novel Coronavirus Infected Pneumonia (NICP). The typical symptom of NCIP patients are persistent fever (≥38 °C), cough and short of breath. The imaging examination showed that the bilateral pneumonia, and multiple mottling and ground-glass opacity. Laboratory tests detection indicated that declined white blood cells and lymphocytes level in the patients [1,2].

The altered serum lipid concentrations, especially cholesterol concentration, have been reported to occur during infection with viruses including human immunodeficiency virus (HIV) and hepatitis C virus (HCV). Some proteins, such as scavenger receptor class B type I (SR-BI), a major receptor of high-density lipoprotein (HDL) and play a crucial role in cholesterol homeostasis, were indicated to involve in HCV infection [3]. Moreover, membrane cholesterol has been indicated to be important component for the entering into host cells of pathogenic viruses [4].

2. Method

2.1. Patients

We recruited 114 COVID-19-infected patients from Jan 2nd to Feb 20th at Wenzhou Central Hospital, in Wenzhou, China. All cases were diagnosed according to the positive result of viral nucleic acid assay by a real-time reverse transcription-polymerase chain reaction (RT-PCR) assay (Bio-germ) in testing respiratory specimens. To avoid the

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Table 1
Serum lipid parameters of patients with COVID-19 on admission.

	Median (IQR) All patients (n = 114)	Controls (n = 80)	*P* value
Age, Median (IQR), Range, y	48.5 (40.8–57.0), 20–93	44.0(36.0–57.8), 21–90	NS
Sex			NS
Male (%)	60 (52.6%)	42 (52.5%)	
Female (%)	54 (47.4%)	38 (47.5%)	
Serum lipid			
Total cholesterol (mmol/l)	3.89 (2.34–4.85)	4.83 (4.27–4.83)	< 0.001
Triglyceride (mmol/l)	1.16 (0.92–1.21)	2.12(0.60–2.36)	0.021
HDL (mmol/l)	1.08 (0.93–1.08)	1.27(1.21–1.41)	< 0.001
LDL (mmol/l)	2.19 (0.94–2.73)	3.06 (2.77–3.06)	< 0.001

Data are median (IQR), n (%), or n/N, where N is the total number of patients with available data. *P* values comparing patients and controls are from $\chi 2$, or Mann-Whitney *U* test.

Table 2
Serum lipid parameters of common group and severe group on admission.

	Median (IQR) Common (n = 87)	Severe (n = 27)	*P* value
Age, Median (IQR), Range, years	46.0 (36.0–54.0), 20–84	62.0 (53.0–71.0), 40–93	< 0.001
Sex			NS
Male (%)	42 (48.3%)	18 (66.7%)	
Female (%)	45 (51.7%)	9 (33.3%)	
Serum lipid			
Total cholesterol (mmol/l)	3.73 (3.31–4.19)	3.82 (3.44–4.58)	NS
Triglyceride (mmol/l)	1.13 (0.93–1.74)	1.32 (1.06–1.63)	NS
HDL (mmol/l)	**1.21 (1.02–1.48)**	**1.01 (0.88–1.20)**	**< 0.001**
LDL (mmol per L)	1.81 (1.52–2.32)	1.88 (1.47–2.28)	NS

Data are median (IQR), n (%), or n/N, where N is the total number of patients with available data. *P* values comparing patients and controls are from $\chi 2$, or Mann-Whitney *U* test.

interference of antiviral treatment on the serum lipid concentrations, we collected the first laboratory results of patients on admission, all of which were untreated at that time. The common or severe cases were diagnosed according to the Novel Coronavirus Pneumonia Diagnosis and Treatment Intern Guidance (the 7th revised version) [5].

For comparative study, we also recruited 80 age-matched healthy controls in healthy center of Wenzhou Central Hospital, excluding the subjects with dyslipidemia, diabetes, cardiovascular diseases and hypertension. All of the health controls had the negative results in the COVID-19 nucleic acid tests. Our study was authorized by Ethics Commission of Wenzhou central hospital (No. L2020-01-003).

To investigate the relationship between lipid profile and severity of COVID-19 infection, we continuously monitored the serum biochemical indexes, blood cell counting, COVID-19 nucleic acid from sputum and chest CT imaging examination of a severe case with interferon-a inhalation treatment from day 1 to day 16 on admission, and additionally measured the indexes on day 30, day 45, and day 80 when she was cured.

2.2. Laboratory tests

For detecting serum lipid concentration, blood samples were collected from each subject after at least 12 h of overnight fasting. For avoiding the interference of treatment to serum lipid concentration, the first laboratory data on admission were collected for our study. Hematological parameters were analyzed on BC-5380 automatic blood cells analyzer. Biochemical parameters were performed on AU5800 chemistry analyzer. All laboratory assays had completed the standardization and certification program.

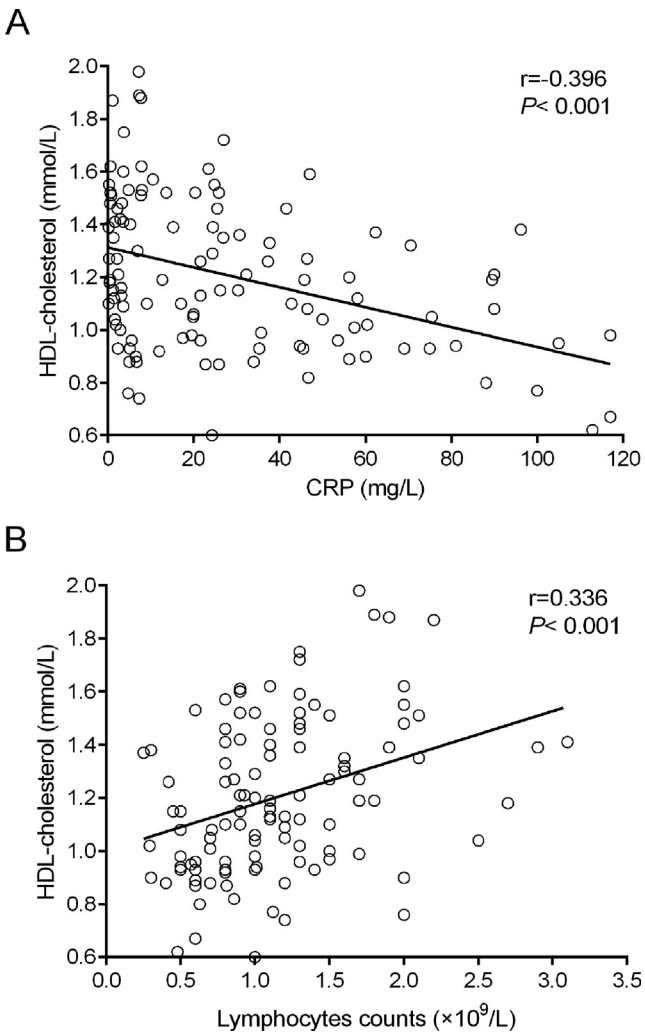

Fig. 1. Correlation analysis of HDL with CRP and Lymphocytes. A: Correlation curve of HDL-cholesterol and CRP; B: Correlation curve of HDL-cholesterol and lymphocyte counts.

2.3. Statistical analysis

Continuous variables were expressed as median (IQR) and analyzed with Mann-Whitney *U* test; categorical variables were expressed as number (%) and compared by χ^2 test between patients and controls groups, or between common group and severe group of patients. The associations between serum HDL-cholesterol and severe COVID-19 infection were determined using binary logistic regression model. Bivariate correlation analysis (Pearson correlation) was performed for analyzing the correlation of serum lipid concentration and other laboratory parameters. A $p < 0.05$ was considered statistically significant. Statistical analyses were performed using the SPSS 17.0 statistical package and GraphPad Prime 6.

3. Results

3.1. Serum lipid concentrations generally decreased at the initial stage of infection

Compared with the heathy controls, the infected patients presented sharply decreased concentrations of serum total cholesterol, HDL-cholesterol and LDL-cholesterol, which were 4.83 (4.27–4.83) *vs* 3.89

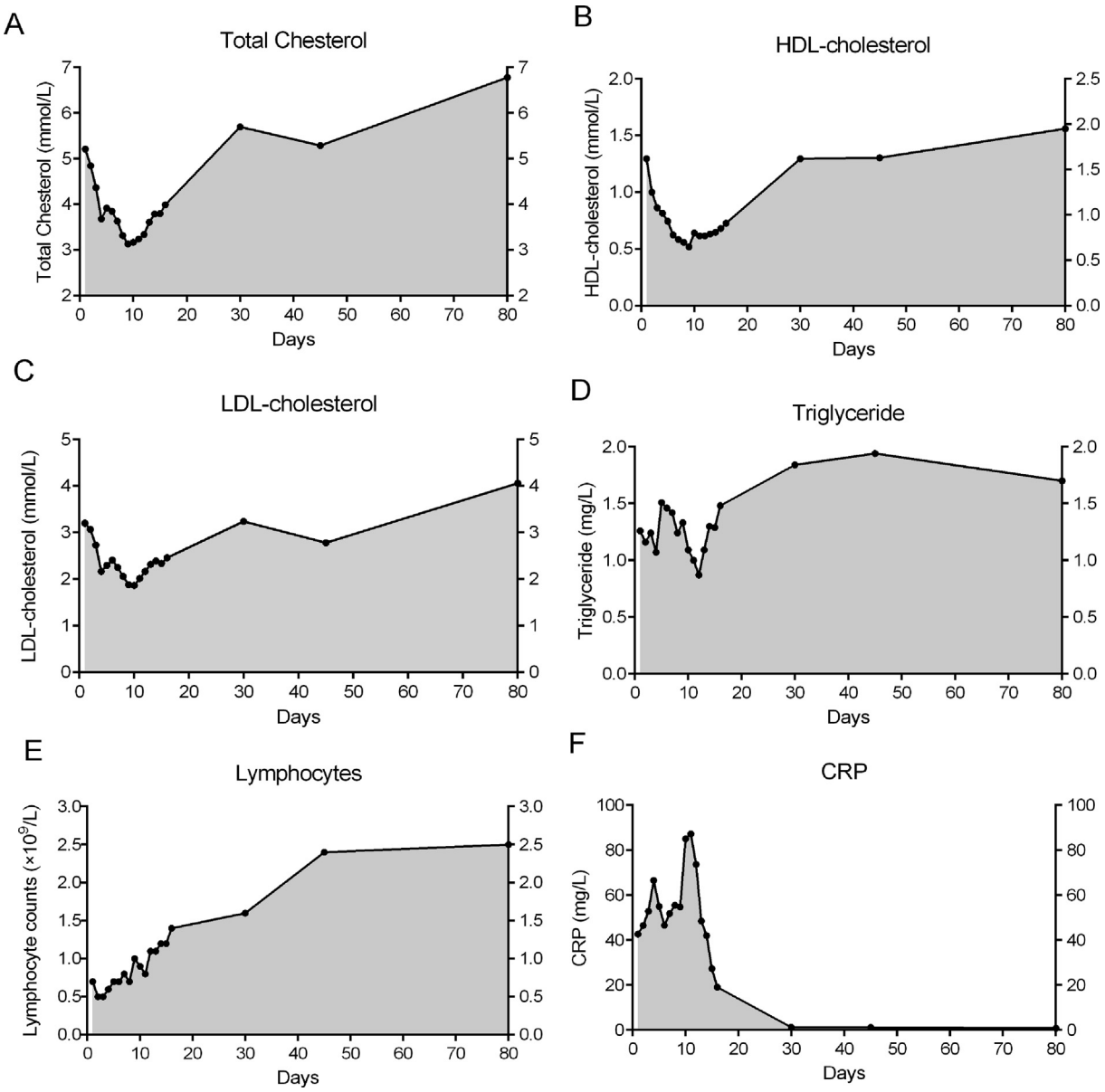

Fig. 2. Laboratory indicators continuously monitored in a severe patient. A-D: Serum lipid profile from day 1 to day 80; E: Blood lymphocyte counts from day1 to day 80; F: Serum CRP from day 1 to day 80.

(2.34–4.85) mmol/l, 1.27(1.21–1.41) *vs* 1.08 (0.93–1.08) mmol/l and 3.06 (2.77–3.06) *vs* 2.19 (0.94–2.73) mmol/l respectively ($P < 0.001$). There was no significant difference in serum triglyceride between the 2 groups (Table 1).

For investigating the relationship between serum lipid and severity of infection, firstly the cases were divided into common group and severe group. As the data showed in Table 2, the average age of the 27 severe patients was 62 y, significantly higher than the average age, 46 y, of the common cases. More notably, median concentration of serum HDL-cholesterol, 1.01 (0.88–1.20) mmol/l, in the severe patients was lower than that, 1.21 (1.02–1.48) mmol/l, in common groups. While there were no differences in serum total cholesterol, triglyceride and LDL-cholesterol concentration between the two groups.

3.2. HDL-cholesterol is correlated with CRP and lymphocyte counts

In correlation analysis, HDL-cholesterol was negatively correlated with CRP ($r = -0.396$, $P < 0.001$, Fig. 1A) and positively correlated with lymphocytes ($r = 0.336$, $P < 0.001$, Fig. 1B).

3.3. The serum lipid changes with the prognosis of the infection in a severe case

As the Fig. 2A–C showed, the severe patient's serum total cholesterol, HDL-cholesterol and LDL-cholesterol concentration dropped persistently until day 9, and then the concentrations were starting recover until the day 16, when she was discharged with negative result of virus nuclear test. On day 30, the total cholesterol, HDL-cholesterol and LDL-cholesterol concentration return to higher concentration, 5.70, 1.62 and 3.24 mmol/l respectively. On day 80, cholesterol increased into 6.78, 1.95 and 4.06 mmol/l (Fig. 2A–C). In this process, HDL-cholesterol concentration in particular showed a change from falling in the early infection to slowly recover as the disease progresses. Additionally, lymphocytes counting presented that gradually rise in this process (Fig. 2E), however, the curves of serum triglyceride and CRP indicated the significant instability of changes (Fig. 2D and F).

Meanwhile, the COVID-19 nucleic acid tests showed the positive results from the day 1 to day 6 on admission. Since then, all of the nucleic acid tests were negative (Fig. 3A). The imaginations of the chest

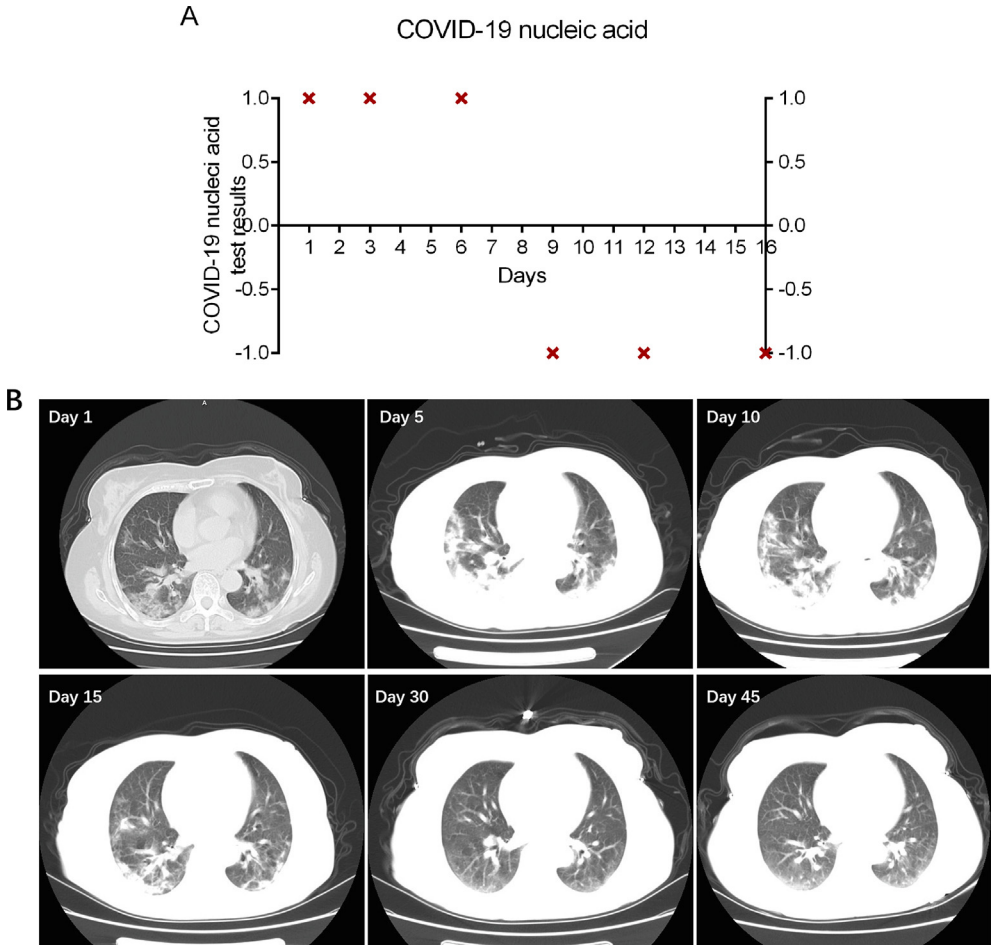

Fig. 3. COVID-19 nucleic acid test and chest CT scans of the severe patient. A: COVID-19 nucleic acid test results from day 1 to day 16, "1" means positive result, "−1" means negative result in PCR.; B: Chest CT scans of the severe patient from day 1 to day 45.

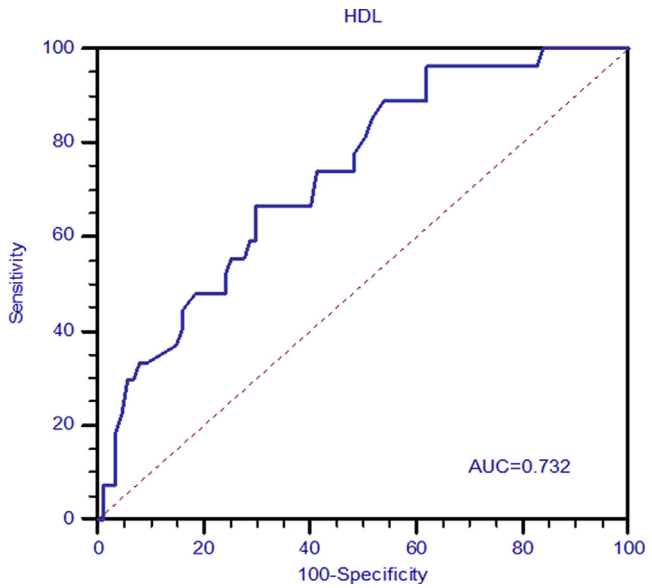

Fig. 4. ROC curves of serum HDL-cholesterol in prediction of severe COVID-19 infection.

CT scan showed the multiple mottling and ground-glass opacity of the both lung in the early stages of the infection, and then turned to be normal as the disease recovered (Fig. 3B).

3.4. Association of serum HDL-cholesterol concentration in COVID-19 infection

Receiver operating characteristic (ROC) analysis was performed to evaluate the ability of serum HDL-cholesterol to predict the severity of COVID-19 infection. As ROC curve showed, AUC was 0.732 (Fig. 4, $P < 0.001$). In further analysis, odd ratio (OR) firstly was used to evaluate the association of serum lipid in COVID-19 infection. Only HDL-cholesterol [adjusted OR 0.023, 95% CI (0.002–0.227)] was independently associated with sever COVID-19 infection (Table 3, $P < 0.001$).

4. Discussion

In this study, we collected 114 COVID-19-infected patients, including 87 common cases and 27 severe cases, and 80 age-matched healthy controls. Our findings indicated significantly dyslipidemia in COVID-19-infected patients. The characteristics of serum lipid profile of COVID-19-infected patients presented declined concentration of total cholesterol, HDL-cholesterol and LDL- cholesterol (Table 1). More notably, the severe cases showed significantly lower concentration of HDL-cholesterol than the common cases (Table 2). Those results are similar to the data in recent clinical studies [6,7].

Further, the correlation between HDL-cholesterol and infection progression was explored. CRP is commonly recognized and sensitive biomarker of inflammation. The result showed that HDL-cholesterol concentration was negatively related with CRP concentration. The correlation between CRP and HDL-cholesterol had been discussed in the

Table 3
Association between HDL and severe COVID-19 infection.

	Unadjusted ORs		Adjusted ORs	
	OR (95%CI)	*P* value	OR*(95%CI)	*P* value
HDL	0.032(0.004–0.234)	0.001	0.023(0.002–0.227)	0.001

* Adjusted: age.

two prospective population-based cohort studies previously. The data in the study showed that the individuals with HDL cholesterol below 0.8 mmol/l (31 mg/dL), CRP was 1.8 (1.2–3.2) and 2.4 (1.5–4.8) mg/l respectively [8], which also indicated that the negatively relationship between HDL-cholesterol and CRP. Moreover, the emerging events recently suggested that the increased CRP concentration was an accurate indicator in predicting COVID-19 infection [9,10]. Our result is consistent with these findings. In addition, we found HDL-cholesterol concentration was positively correlated with lymphocytes count. In the previous reports, the declined lymphocytes counting was found in the most patients and considered to be a predictor in COVID-19 infection [9–11]. Laboratory studies also suggested that COVID-19 might mainly act on lymphocytes, especially T lymphocytes, ant the substantial decrease in lymphocytes counting indicated COVID-19 might inhibits cellular immune function [1].

In terms of the findings in a severe patient we constantly monitored, the concentration of serum total cholesterol, HDL-cholesterol and LDL-cholesterol, as well as lymphocytes counting in the process of infection were able to effectively reflect the process of the disease, however, HDL-cholesterol concentration was especially significant (Fig. 2). Notably, the changes of serum HDL-cholesterol were consistent with that of chest CT imaging examination and virus nucleic acid tests (Fig. 3A and B), which are indicators associated with disease prognosis.

All of the above results suggested that HDL-cholesterol concentration might be closely associated with the progression and severity of COVID-19 infection. In our further analysis, ROC curve (AUC = 0.732, Fig. 4) and adjusted OR value (0.023, Table 3) indicated that the value of HDL-cholesterol as an assessment of the severity of COVID-19 infection was appropriate.

Currently, COVID-19 infection-associated dyslipidemia had been reported in several clinical studies [6,7,12], however, underlying mechanism of cholesterol metabolism in COVID-19 infection is still unknow. Hitherto, the most widely investigation of virus infection with dyslipidemia is HIV-associated dyslipidemia [13,14]. In HIV-1 infection, low serum/plasma HDL-cholesterol was confirmed to be associated with the impairment of ATP-binding cassette transporter A1-dependent cholesterol efflux from macrophages, and the activation of endothelial lipase and phospholipase A2 by inflammation [15,16].

Although it is unclear whether declined serum HDL-cholesterol in COVID-19 infection shares the same mechanism as HIV-1 infection, HDL particles and cholesterol were indicated to be closely related to virus infection. The studies *in vitro* showed that cholesterol on lipid rafts was proved to be required for the early stage of SARS-CoV replication and during the binding stage of SARS-CoV entering host cells [17,18]. Furthermore, HDL generally is an anti-inflammatory lipoprotein [19–21]. However, inflammation was reported to leads to structural alterations of HDL particles and caused the accumulation of the acute phase protein serum amyloid A (SAA) within the protein moiety of HDL [22]. SAA-enriched HDL particles lose the anti-inflammatory properties and even promoted the pro-inflammatory activation of macrophages and translocation of TLR4 into lipid rafts [23]. In view of these findings, we speculated that HDL lost its anti-inflammatory properties, as well as consumed cholesterol during the COVID-19 infection. It might be explained the significantly dropped HDL-cholesterol concentration in the severe COVID-19-infected patients. It needs to be verified by further studies.

Our study has several limitations. As a retrospective study, our number of samples is small, as well as from single medical center, which might induce the deviation of the results. Additionally, the widely used antiviral treatment, such as Lopinavir, had been confirmed to lead to dyslipidemia. In contrast, interferon has little effect on lipid metabolism. Unfortunately, most of the patients were treated with Lopinavir, we only collected one case with interferon-a treatment and monitored the case constantly. If there are more, our conclusion will be more convincing.

Taken together, the patients with COVID-19 infection showed that serum HDL-cholesterol concentration significantly declined in the early stage of the disease, especially in those severe infected patients. Based on our current analysis, decreased serum HDL-cholesterol is associated with the severity of COVID-19 infection.

CRediT authorship contribution statement

Xingzhong Hu: Conceptualization. **Dong Chen:** Project administration. **Lianpeng Wu:** Methodology. **Guiqing He:** Data curation. **Wei Ye:** Writing - original draft.

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