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# Analysis of adjunctive serological detection to nucleic acid test for severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection diagnosis

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## ABSTRACT

**Background:** The severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) caused coronavirus disease 2019 (COVID-19) epidemic in China, December 2019. The clinical features and treatment of COVID-19 patients remain largely elusive. However, accurate detection is required for SARS-CoV-2 infection diagnosis. We aimed to evaluate the antibodies-based test and nucleic acid-based test for SARS-CoV-2-infected patients.

**Methods:** We retrospectively studied 133 patients diagnosed with SARS-CoV-2 and admitted to Renmin Hospital of Wuhan University, China, from January 23 to March 1, 2020. Demographic data, clinical records, laboratory tests, and outcomes were collected. Data were accessed by SARS-CoV-2 IgM-IgG antibody test and real-time reverse transcriptase PCR (RT-PCR) detection for SARS-CoV-2 nucleic acid in COVID-19 patients.

**Results:** Of 133 COVID-19 patients, there were 44 moderate cases, 52 severe cases, and 37 critical cases with no differences in gender and age among three subgroups. In RT-PCR detection, the positive rate was 65.9%, 71.2%, and 67.6% in moderate, severe, and critical cases, respectively. Whereas the positive rate of IgM/IgG antibody detection in patients was 79.5%/93.2%, 82.7%/100%, and 73.0%/97.3% in moderate, severe, and critical cases, respectively. Moreover, the IgM and IgG antibodies concentrations were also examined with no differences among three subgroups.

**Conclusion:** The IgM-IgG antibody test exhibited a useful adjunct to RT-PCR detection, and improved the accuracy in COVID-19 diagnosis regardless of the severity of illness, which provides an effective complement to the false-negative results from a nucleic acid test for SARS-CoV-2 infection diagnosis after onsets.

## 1. Introduction

The novel severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) causing coronavirus disease 2019 (COVID-19) emerged in China in later December 2019, and has become pandemic after global spreading. SARS-CoV-2 has infected 76,819 people out of which 12,077 were critical, 2,251 died (2.9% fatality rate), and 18,878 clinically

recovered during the first 50 days of the outbreak in China [1,2]. As of June 6, 2020, it has been reported 6,663,304 confirmed cases and 392,802 deaths by SARS-CoV-2 infection among over 200 countries and territories [3]. To mitigate the risk of spread it is necessary to investigate and develop effective treatment and diagnostic options. The signs and symptoms of SARS-CoV-2 infection are not specific, most are associated with respiratory complications after onsets such as cough

**Abbreviations:** RT-PCR, real-time reverse transcriptase polymerase chain reaction; COVID-19, coronavirus disease 2019; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; IgM, immunoglobulin M; IgG, immunoglobulin G; CT, Computed Tomography; ICU, intensive care unit; RLU, relative light unit; antibody unit, AU; MERS, Middle East respiratory syndrome coronavirus; d.p.o, days post-disease onset; ddPCR, droplet digital PCR; CRISPR, Clustered regularly interspaced short palindromic repeats; mNGS, metagenomic next-generation sequencing

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dyspnea, and viral pneumonia, but the mortality of critically ill patients with SARS-CoV-2 pneumonia is also considerable [4,5]. Therefore, specific COVID-19 diagnostic tests are required to confirm suspected cases accurately and shortly. Besides specific diagnostic techniques, appropriate samples or specimens for the detection of the viral genome are also of high importance [6,7].

Previous studies on COVID-19 pneumonia have largely focused on clinical characteristics and epidemiology [8,9]. However, very limited details are available related to effective diagnostic strategies. In the current situation, the specificity and sensitivity of the tests are not widely known, therefore, testing of multiple specimen types is recommended [10,11]. The most widely used tests in the current situation are based on nucleic acid detection and antibody detection. Although the viral nucleic acid RT-PCR test has become the standard method for SARS-CoV-2 infection diagnosis, high false-negative rates were reported [12]. Thus, an accurate and infallible detection for SARS-CoV-2 infection diagnosis is urgently needed.

Upon coronavirus infection, IgM antibodies are produced as an early immune response after infection in the body, which may indicate current infection or new infection. During the early stage of the infection (days 4–10), the IgM component of the test provides a sensitivity of just 70%. This rises rapidly to 92.3% between days 11 and 24, and the IgG component of the test offers a sensitivity of 98.6% during this phase of the infection or even longer [13]. IgG antibodies are the main antibodies produced as an immune response, indicating that the disease has entered a recovery period or that there is a prior infection [14,15]. Therefore, combined tests of immunoglobulin M (IgM) and immunoglobulin G (IgG) antibodies can not only provide the diagnosis of infectious diseases but also help to evaluate the stages of infection in the body [12,13].

To explore the accurate and reliable detection for COVID-19 diagnosis, the present work was conducted to evaluate the nucleic acid-based and antibodies-based tests for SARS-CoV-2-infected patients. A total of 133 clinical nasopharyngeal swabs and serum samples from confirmed COVID-19 patients were divided into three subgroups by the severity of illness and surveyed with nucleic acid test and IgM-IgG antibody test, respectively. Moreover, the IgM and IgG antibodies concentrations were also investigated with no differences among three subgroups based on the severity of COVID-19. Our findings revealed that the adjunct of the serological test to nucleic acid test improves the accuracy in COVID-19 diagnosis after onsets.

## 2. Materials and methods

### 2.1. Patients

A total number of 133 patients diagnosed with SARS-CoV-2 infection in Renmin Hospital (Wuhan University, China) from January 23 to March 1, 2020, were included as the case group in this study. All patients were diagnosed according to the “pneumonia diagnosis protocol for novel coronavirus infection (trial version 5)”, subjected to the tests including clinical examination, Computed Tomography (CT), and real-time reverse-transcription polymerase chain reaction (RT-PCR) for SARS-CoV-2. The SARS-CoV-2 group was divided into three subgroups according to new pneumonia diagnosis and treatment of COVID-19 (trial version 5), including 44 moderate cases, 52 severe cases, and 37 critical cases (Table 2).

The moderate, severe, and critical cases were defined based on symptoms severity and condition of the patients according to WHO interim guidance [16]. Briefly, moderate cases were defined as mild symptoms including fever, cough, headache, or soreness from cough but no pain, while severe cases as severe signs including inflammation of lungs, extreme breathlessness, pain in the chest, fast heartbeat, or unwell appearance and low blood pressure. The critical patients were defined as those admitted to the intensive care unit (ICU) who required mechanical ventilation or had a fraction of inspired oxygen (FiO<sub>2</sub>) of at

least 60% or more as previously described [4].

### 2.2. Data collection

Data on biochemical parameters were obtained from all 133 confirmed SARS-CoV-2 infected patients, which was confirmed by a broad series of investigations including clinical examination, laboratory tests, chest X-rays and two independent real-time reverse-transcription polymerase chain reaction (RT-PCR) for SARS-CoV-2, with SARS-CoV-2 ORF1ab/N PCR detection kit (GeneDx Biotech, Shanghai, China), as well as using a SARS-CoV-2 antibody detection kit (YHLO Biotech, Shenzhen, China). According to the standard procedure protocol, the RT-PCR test was performed for SARS CoV-2 nucleic acid of nasopharyngeal swabs. A cycle threshold (Ct) value less than 37 was defined as a positive test result, and a Ct value of 40 or more was defined as a negative test according to the previous definition [9]. The IgM-IgG antibody test was performed in serum or plasma as previously reported [17]. Clinical details and laboratory results were collected during routine clinical work through the patient's interview. The study was approved by the Ethics Committee and Institutional Review Board of the Renmin Hospital of Wuhan University (certificate no. WDRY2020-K066).

### 2.3. Nucleic acid test

Fluorescent quantitative RT-PCR was used to detect ORF1ab and the nucleocapsid protein N genes in the SARS-CoV-2 genome. SARS-CoV-2 nucleic acid test results from Ct (Cycle threshold) value interpretation was subject to the manufacturer's specification, and the suspected results were notified of clinical resampling review. The laboratory test results of both positive SARS-CoV-2 ORF1ab and N genes, or two consecutively positive SARS-CoV-2 ORF1ab gene or N gene can determine the presence of SARS-CoV-2 nucleic acid in the samples.

### 2.4. Serological test

SARS-CoV-2 IgM-IgG antibody detection kits were adopted for the direct chemiluminescence technique of two-step indirect immunoassay, which were used for the qualitative detection of SARS-CoV-2 in human serum or plasma. The procedure was performed according to the kit protocol provided, and the test results were indicated by luminous strength (relative light unit, RLU). The concentrations (AU/ml) of SARS-CoV-2 IgM or IgG antibodies in the sample was correlated with the RLU, and calculated based on the RLU and built-in calibration curve, with the value of AU/ml > 10 as the positive reaction.

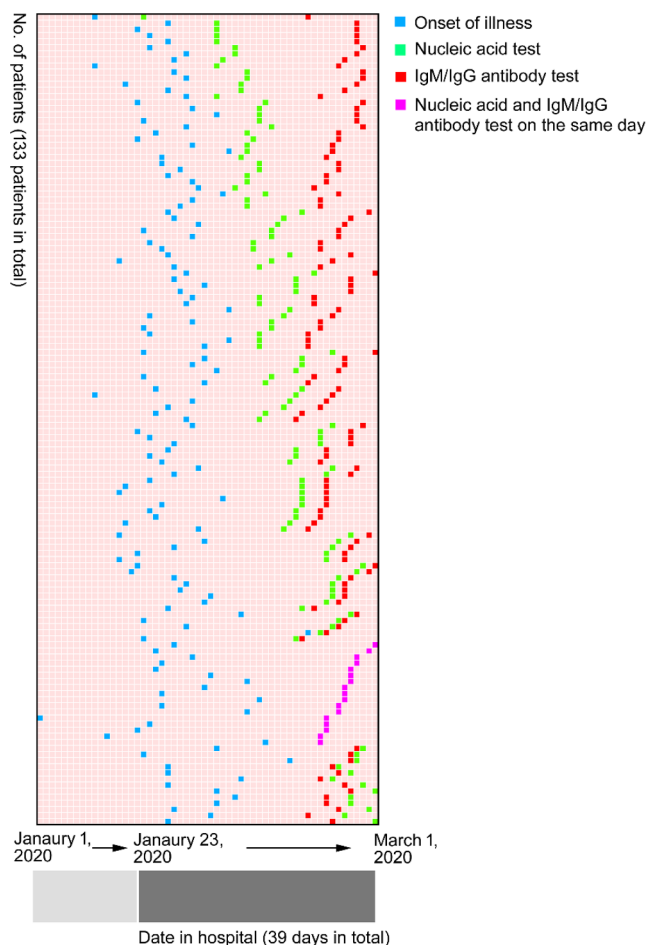
### 2.5. Statistical analysis

SPSS software version 25.0 (IBM Corp.; Armonk, NY, USA) was used for statistical analysis. All quantitative data in non-normal or unknown distribution were expressed as median and interquartile range. Kruskal-Wallis test was used to analyze differences among groups for the measurement data that did not meet the normal distribution. The Chi-square ( $\chi^2$ ) test was used for the difference between groups of enumeration data. In all tests,  $P < 0.05$  was defined as statistically significant

## 3. Results

### 3.1. The value of antibody and RT-PCR detection for SARS-CoV-2 infection diagnosis

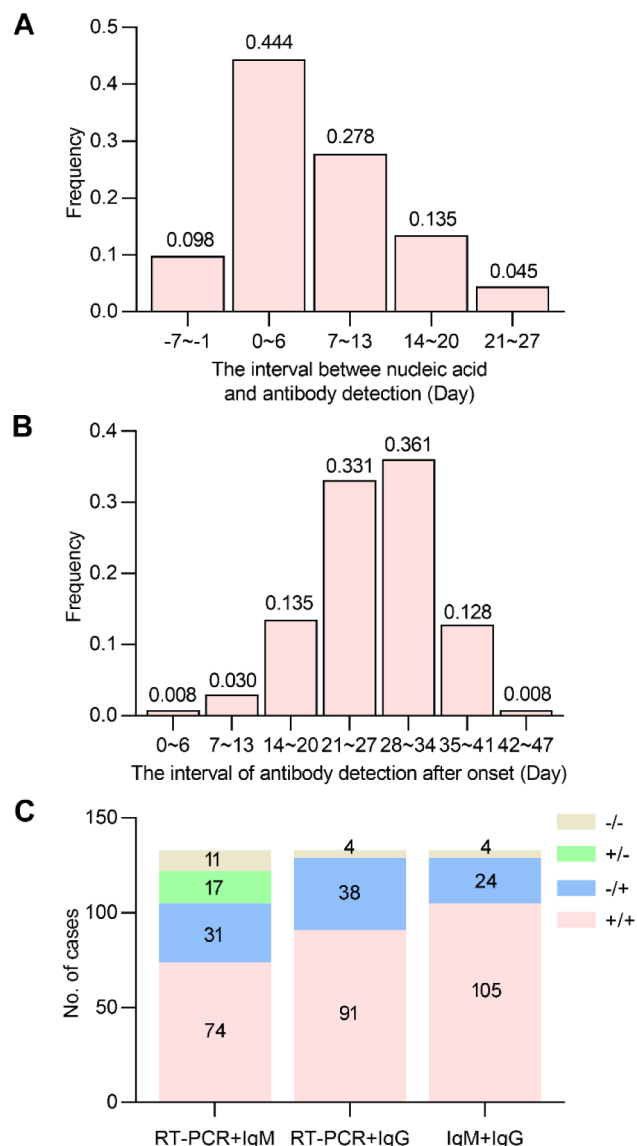
Of 133 COVID-19 patients admitted in the designated hospital between January 23 and March 1, 2020, RT-PCR and IgM-IgG antibody tests were successively performed in the diagnosis of SARS-CoV-2 infection (Fig. 1). Detailly, 44.4% of patients were tested for nucleic acid



**Fig. 1.** The time features of nucleic acid test and IgM-IgG antibody test for COVID-19 patients. The graticule represents the panoramic timeline of onset of illness (blue grid), nucleic acid test (green grid), and IgM-IgG antibody test (red grid) of 133 patients infected with SARS-CoV-2. The interval of data from January 23, 2020, to March 1, 2020, was the period of hospitalization. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

and IgM-IgG antibody successively within a week, while 45.8% of patients were diagnosed with the IgM-IgG antibody test a week or even longer after nucleic acid test (Fig. 2A). To further examine the interval of IgM-IgG antibody test after COVID-19 patients' onsets, 82.7% of patients were tested for IgM-IgG antibody during 14–35 days after onset, while 13.6% of patients were tested more than 35 days after onset, only 3.8% of patients were tested within 14 days after onset (Fig. 2B).

It was illustrated that the positive rate was 78.9% (105/133) and 97.0% (129/133) in IgM and IgG antibody test, respectively, while 68.4% (91/133) in RT-PCR detection for SARS-CoV-2 infection in all COVID-19 patients (Table 1). In detail, 31 patients were tested IgM positive and RT-PCR negative, with the RT-PCR missed diagnosis rate of 31.6% (42/133), while 17 patients were tested IgM negative and RT-PCR positive, with the IgM missed diagnosis rate of 21.1% (28/133). Nevertheless, only 11 patients were tested the combined RT-PCR and IgM both negative, with the missed diagnosis rate of 8.3% (11/133) (Fig. 2C, subset 1). In a similar analysis, 38 patients were tested IgG positive and RT-PCR negative, with the RT-PCR missed diagnosis rate of 31.6% (42/133) whereas the IgG missed diagnosis rate of 3.0% (4/133). In the combined RT-PCR and IgG tests, only 4 patients were both negative with the missed diagnosis rate of 3.0% (4/133) (Fig. 2C, subset 2), which significantly decreased false-negative results in COVID-19 diagnosis (Table 1).



**Fig. 2.** Nucleic acid and antibody tests in patients with COVID-19. (A) The histogram of frequency distribution reflects that 44.4% of patients were tested for nucleic acid and IgM-IgG antibody successively within a week, while 45.8% of patients were tested for IgM-IgG antibody after a week or even longer. The number in the horizontal axis indicated the interval days of IgM-IgG antibody test after nucleic acid test. (B) The histogram of frequency distribution of interval of IgM-IgG antibody test after onset of illness since January 1, 2020. 82.7% of patients were tested for IgM-IgG antibody during 14–35 days after onset, while 13.6% of patients were tested more than 35 days after onset, and 3.8% of patients were tested within 14 days after onset. (C) The distribution of cases number from results of different tests in COVID-19 diagnosis by three subsets, 1: RT-PCR and IgM, 2: RT-PCR and IgG, and 3: IgM and IgG. + and - stand for positive and negative results in the indicated tests, respectively. No., Number.

We also observed IgM and IgG positive rates were 78.9% (105/133) and 97.0% (129/133), respectively, with the missed diagnosis rate of 3.0% (4/133) in IgM-IgG antibody detection (Fig. 2C, subset 3). Of note, the 4 patients with both antibody and RT-PCR detection negative may be related to the improvement and recovery with clinical treatments. Altogether, these data suggested that the antibody test could be an effective supplement to RT-PCR detection in the diagnosis of SARS-CoV-2 infection.

**Table 1**  
The detection of antibodies and RT-PCR tests for SARS-CoV-2 infection diagnosis.

Antibodies against SARS-CoV-2		SARS-CoV-2 RNA		Sample Quantity
		+	-	
IgM	+	74	31	105
	-	17	11	28
	Total	91	42	133 <sup>a</sup>
IgG	+	91	38	129
	-	0	4	4
	Total	91	42	133 <sup>b</sup>

Note: + stands for positive, while - stands for negative. <sup>a</sup>,  $P = 0.059 > 0.05$ ; <sup>b</sup>,  $P < 0.001$ .  $P < 0.05$  indicates statistically significant difference between the two detection methods.

**3.2. The value of RT-PCR and IgM-IgG antibody tests in COVID-19 patients in different subgroups**

Considering the severity of COVID-19 patients from the critical care resources in hospitals [4], the COVID-19 patients were divided into three subgroups for further analysis. The three subgroups were divided as 44 moderate cases (22 males and 22 females, the median age was 67.5 [64–71.75]), 52 severe cases (28 males and 24 females, the median age was 68 [61.25–74]), and 37 critical cases (20 males and 17 females, the median age was 70 [60–76.5]) (Table 2). There was no significant difference in gender and age among the three subgroups.

In the RT-PCR detection for viral RNA in three subgroups of COVID-19 patients, the positive rate was 65.9% in moderate cases, 71.2% in severe cases, and 67.6% in critical cases, respectively (Table 3). However, we didn't observe significant differences in positive rate among three subgroups of COVID-19 patients ( $P > 0.05$ ).

Furthermore, the antibodies-based tests were performed in three subgroups of COVID-19 patients. In IgM antibody detection in patients infected with SARS-CoV-2, the positive rate was 79.5% in moderate cases, 82.7% in severe cases, and 73.0% in critical cases, respectively. Similarly, the positive rate from IgG antibody test was 93.2% in moderate cases, 100% in severe cases, and 97.3% in critical cases, respectively (Table 3). There was no statistically significant difference in the positive rate of IgM-IgG antibody detection for COVID-19 patients among the three subgroups ( $P > 0.05$ ). In total, it appeared an increased positive rate in antibodies-based tests to that in the nucleic acid test, indicating that the adjunct of serological test improved the accuracy in the COVID-19 diagnosis regardless of the severity of illness.

**3.3. The concentrations of IgM-IgG antibody detection for COVID-19 patients in different subgroups**

Finally, the concentrations of IgM and IgG antibodies against SARS-CoV-2 in the serological tests for COVID-19 patients in different subgroups were examined. The concentration of IgM in patients was 29.19

**Table 2**  
Demographic and baseline characteristics of COVID-19 patients in different subgroups<sup>a</sup>.

	Moderate (n = 44)	Severe (n = 52)	Critical (n = 37)	P value
Age (Year)	67.5 (64–71.75)	68 (61.25–74)	70 (60–76.5)	0.889
Gender (Male/ Female)	22/22	28/24	20/17	0.913

Note: <sup>a</sup> All patients were divided into three subgroups based on the severity of COVID-19, namely mild cases, moderate cases, and critical cases. Median and interquartile range of age were listed.

AU/ml [17.04–61.02] in moderate cases, 40.76 AU/ml [13.56–90.13] in severe cases, and 23.25 AU/ml [8.67–104.5] in critical cases, respectively. Meanwhile, the concentration of IgG in patients was 147.73 AU/ml [89.53–171.6] in moderate cases, 148.63 AU/ml [130.95–167.7] in severe cases, and 140.4 AU/ml [93.79–162.8] in critical cases, respectively (Table 4). Collectively, there were no significant differences in the concentrations of IgM and IgG antibodies among three subgroups of COVID-19 patients, suggesting the serological test behaved as a considerable diagnosis for COVID-19 patients regardless of the severity of illness.

**4. Discussion**

The outbreak of pneumonia caused by SARS-CoV-2 spreads rapidly, posing a serious threat to the lives and health of the people, which has become a serious global issue. SARS-CoV-2 belongs to the coronavirus beta genus, with a linear single-stranded positive RNA, the seventh coronavirus known to infect humans after SARS (2002) and MERS (Middle East respiratory syndrome coronavirus) (2012) [18]. There are various assays developed to detect different regions of SARS-CoV-2 genome using RT-PCR [10,19]. In the present study, we evaluated both antibody and nucleic acid -based diagnostic strategies on suspected patients with moderate to critical symptoms for COVID-19. Of the total 133 patients were tested, 68.4% (91/133) were positive in the case of RT-PCR and 78.9% (105/133) in the case of the antibody test. It was also observed an increased positive rate in antibodies-based tests to that in nucleic acid test in the diagnosis for COVID-19 patients in different subgroups (moderate cases, severe cases, and critical cases). Our findings suggested that the IgM-IgG antibody test provides an effective complement to the false-negative results from nucleic acid test for COVID-19 diagnosis.

Recently, chest CT scans were applied for the rapid detection of SARS-CoV-2 induced COVID-19 [11,20]. The chest x-ray or chest CT provides more information, but these are not conclusive as not all the patients with COVID-19 developed pneumonia and might produce false results as many other things can also cause pneumonia [21,22]. Therefore, a more effective strategy such that testing antibodies or RNA is important. The conventional serologic assays, droplet digital (dd) PCR, CRISPR-based, and metagenomic next-generation sequencing (mNGS) techniques are also novel approaches for the detection of SARS-CoV-2. In fact, the optimal diagnosis ways for SARS-CoV-2 are usually selected based on the periods of illness onsets (eg. RT-PCR or serologic assays), the viral load of specimens (eg. RT-PCR or ddPCR assays), and the aim of pathogen identification of unexplained pneumonia (eg. CRISPR-based or mNGS techniques) [12,23–25]. Hence, it is highlighted that the combined tests on SARS-CoV-2 antibodies and RNA for the high accuracy of COVID-19 diagnosis according to the desired requirements. SARS-CoV-2 is an emerging kind of infectious pathogen and the immunological testing reagents have recently been developing [12]. It has been established a high sensitivity and specificity of SARS-CoV-2 IgM and IgG antibodies detection in serum or plasma from COVID-19 patients, without cross-reactivity within samples from non-infected individuals [12,26]. Although the antibodies generated after a period of the onset of infection, their detections side by side with RT-PCR detection were found more promising as an accurate detection strategy in the current situation.

It is suggested that serum antibodies-based tests could be effectively adjunctive to RT-PCR test, particularly for patients who had the substantial duration of illness, in whom RT-PCR may be negative. The combining RT-PCR and IgM-IgG antibody detections significantly improved the sensitivity of pathogenic diagnosis for COVID-19 even in the early phase of one week [27]. Moreover, there is an increase in the positive rate for SARS-CoV-2 IgM and IgG with an increasing number of days post-disease onset (d.p.o). As a result, after 10 to 24 d.p.o or even longer to 35 d.p.o, IgM-IgG antibody detection can be an important supplementary method for COVID-19 diagnosis [26,27]. Serological



**Table 3**  
The RT-PCR detection and IgM-IgG test in COVID-19 patients in different subgroups.

SARS-CoV-2		Moderate (n = 44)		Severe (n = 52)		Critical (n = 37)		$\chi^2$	P value
		No. (+)	Rate (+)	No. (+)	Rate (+)	No. (+)	Rate (+)		
RNA	N	29	65.9%	38	73.1%	25	67.6%	0.636	0.728
	ORF1ab	33	75.0%	42	80.8%	27	73.0%	0.84	0.657
	N&ORF1ab	29	65.9%	37	71.2%	25	67.6%	0.321	0.852
Antibody	IgM	35	79.5%	43	82.7%	27	73.0%	1.243	0.537
	IgG	41	93.2%	52	100%	36	97.3%	3.409	0.137

Note: No., number; Rate (+), positive rate. N and ORF1ab, the nucleocapsid protein N and ORF1ab genes in the SARS-CoV-2 genome.

**Table 4**  
The concentrations of IgM and IgG antibodies against SARS-CoV-2 (AU/ml) in COVID-19 patients in different subgroups.

Antibodies against SARS-CoV-2	Moderate n = 44	Severe n = 52	Critical n = 37	P value
IgM	29.19 (17.04–61.02)	40.76 (13.56–90.13)	23.25 (8.67–104.5)	0.446
IgG	147.73 (89.53–171.6)	148.63 (130.95–167.7)	140.4 (93.79–162.8)	0.182

Note: The concentration unit of antibodies in serum samples is AU/ml. The value of AU/ml > 10 is considered as a positive reaction.

based testing will become a very valuable tool to access previous exposures and prevalence in the population [13]. In our study, we examined the interval of IgM-IgG antibody test after COVID-19 patients' onset, 82.7% of patients were tested for IgM-IgG antibody during 14–35 d.p.o, while 13.6% of patients were tested more than 35 d.p.o, and only 3.8% of patients were tested within 14 d.p.o. We also retrospectively observed that some patients were tested after onsets in a prolonged period, which is explained that the lack of timely RT-PCR and IgM-IgG antibody tests in early COVID-19 epidemic in Wuhan, in January 2020. Fortunately, the status had been improved that portions of patients were timely tested with a reasonable interval after onsets since late January 2020, due to the effective interventions and efforts made by the Chinese government. Nevertheless, the serological test still behaved as a considerable diagnosis for COVID-19 patients after onsets.

Certainly, it was confirmed that the detection sensibility was higher in the IgG-IgM combined antibody test than in individual IgG or IgM antibody test [12,28]. However, a low viral load in patients' throat and the limitation of RT-PCR result in a significant number of false-negative reports, which should not be ignored [25]. In general, the coronavirus stimulates the immune response and IgM antibodies are produced after 4–24 days upon infection and then quickly decline until disappear, while on the other hand, IgG antibodies are usually produced after IgM and continue to rise and remain high in the body for long periods [14,15]. For treatment monitoring and status of the disease, the decrease or even disappearance of the concentration of IgM and the increase in the concentration of IgG indicates the severity of the patient and the immunity to the pathogenicity of SARS-CoV-2. Actually, we found there were no significant differences in the concentrations of IgM and IgG antibodies of COVID-19 patients among three subgroups based on the severity of illness. Therefore, further investigations should be made on a broad range and mainly focus on the antibody's response pattern and severity status of a larger scale of COVID-19 patients on the bases of antibodies production.

In conclusion, our study revealed that the IgM-IgG antibody test exhibited a useful adjunct to RT-PCR detection, which improved the accuracy in the COVID-19 diagnosis regardless of the severity of illness. Considering the significance of this ongoing COVID-19 pandemics, we believe that our findings are important in terms of providing promising diagnostic options based on age or gender groups, as well as the

severity of symptoms. We further recommend the IgM-IgG antibody test provides an effective complement to nucleic acid test for SARS-CoV-2 infection diagnosis.

#### CRediT authorship contribution statement

**Rui Liu:** Data curation, Formal analysis, Methodology. **Xinghui Liu:** Formal analysis, Funding acquisition, Investigation, Methodology. **Li Yuan:** Data curation, Formal analysis. **Huan Han:** Formal analysis, Resources. **Muhammad Adnan Shereen:** Data curation, Formal analysis, Investigation, Methodology. **Jiesheng Zhen:** Methodology, Resources. **Zhili Niu:** Investigation, Methodology, Resources. **Dong Li:** Investigation, Resources. **Fang Liu:** Investigation, Methodology, Supervision, Validation. **Kailang Wu:** Investigation, Supervision, Validation. **Zhen Luo:** Data curation, Funding acquisition, Validation, Writing - original draft. **Chengliang Zhu:** Conceptualization, Funding acquisition, Supervision, Writing - review & editing.

#### Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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#### Appendix A. Supplementary material

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.intimp.2020.106746>.

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