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Frequency of SARS-CoV-2 antecedent infection in patients with Kawasaki disease

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ABSTRACT

Since the COVID-19 pandemic, it has been found that SARS-CoV-2 antecedent infection can cause multisystem inflammatory syndrome in children. However, the frequency of SARS-CoV-2 antecedent infection in Kawasaki disease (KD) is unknown. The purpose of this study was to investigate the frequency of SARS-CoV-2 infection that preceded the onset of KD. This study is a multi-center observational study. The subjects were patients who were diagnosed with KD at 4 hospitals from April 1, 2020 to August 31, 2022. Serum SARS-CoV-2 IgM and total antibody including IgG levels were measured by the chemiluminescence immunoassay method before and after treatment for KD. A total of 55 patients participated in the study. The first antibody measurement before the initial treatment was performed on a median of 4th days of illness and the second antibody measurement was performed after the initial treatment on a median of 12th days of illness. No patient had a significant increase in SARS-CoV-2 IgM levels in two measurements. Only one patient (1.8%) had elevated total antibodies including IgG, and the patient had a history of COVID-19 6 months before the onset of KD. SARS-CoV-2 antecedent infection before the onset of KD was not observed in this study, and strong association between development of KD and SARS-CoV-2 infection was not suggested.

Keywords: antecedent infection, antibody, COVID-19, Kawasaki disease, SARS-CoV-2

Abbreviations:
KD: Kawasaki disease
COVID-19: coronavirus disease 2019
SARS-CoV-2: severe acute respiratory syndrome coronavirus 2
MIS-C: multisystem inflammatory syndrome in children

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INTRODUCTION

Kawasaki disease (KD) is a self-limiting, acute systemic inflammatory vasculitis that occurs mainly in infants and is characterized by its effects on the coronary arteries. Although KD has the highest incidence in Japan, it has been reported in children from various regions and races worldwide.[1] KD aetiology has not been ascertained, and a hypothesis mentions infections, including viral, as the cause.[2] Genetic factors have also been reported to contribute to KD pathogenesis.[3,4]

During the coronavirus disease 2019 (COVID-19) pandemic, there has been a series of reports in Europe and the United States of America of children who developed inflammatory syndromes similar to KD after severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection.[5] This syndrome is now known as multisystem inflammatory syndrome in children (MIS-C). However, there are many differences in symptoms and laboratory findings from typical KD, and it is unclear whether the disease belongs to the same disease spectrum.[6] Contrastly, epidemiological reports show that KD cases have decreased during the COVID-19 pandemic in Japan and other countries and regions.[7,8] Therefore, the relationship between SARS-CoV-2 infection and KD onset remains unclear. This study aimed to investigate the presence and frequency of SARS-CoV-2 infection that preceded KD onset by measuring serum SARS-CoV-2 antibody levels in patients with KD during the COVID-19 pandemic.

MATERIALS AND METHODS

Study design and population

This was a multi-center prospective observational study. The subjects were patients aged <18 years diagnosed with KD, including incomplete type, and treated at four hospitals in Aichi Prefecture, Japan, from 1 April 2020 to 31 August 2022. KD diagnosis was based on the sixth edition of the Diagnostic Guidelines for Kawasaki Disease.[9] Patients with a history of KD or receiving γ globulin, and patients or their guardians who did not give informed consent were excluded.

SARS-CoV-2 antibody test

Serum was collected from patients before starting treatment for KD and on 10th to 14th days of illness, and SARS-CoV-2 antibody levels were measured using two types of reagents. One was a SARS-CoV-2 IgM measurement reagent (Fujirebio, Inc Tokyo, Japan), which detects IgM against the receptor binding domain of SARS-CoV-2 using a chemiluminescent enzyme immunoassay. The SARS-CoV-2 IgM measurement reagent has been reported to be positive in 90 out of 100 patients with confirmed diagnosis of SARS-CoV-2 infection by polymerase chain reaction (PCR) or antigen tests.[10] The other was Elecsys Anti-SARS-CoV-2 (Roche Diagnostics KK Tokyo, Japan), an immunoassay reagent that measures antibodies (including IgG) against the SARS-CoV-2 nucleocapsid using electrochemiluminescence immunoassay. The Elecsys Anti-SARS-CoV-2 has been reported to have a sensitivity of 85.3% 7–13 days after and 99.5% after 14 days or more after a confirmed diagnosis of SARS-CoV-2 by PCR.[11] For both methods, the cutoff index (COI) was calculated as the serum luminescence level divided by the SARS-CoV-2 standard positive solution luminescence level.

Clinical data

Demographic data such as height, weight, sex, presence or absence of each major symptom of KD, and laboratory examination results were obtained from the medical records of all participants.

Kobayashi score, which predicts intravenous high-dose immunoglobulin (IVIG) resistance,[12] was calculated. In addition, information was obtained on the presence or absence of residual coronary artery complications within 4 weeks after the onset of KD and other complications. Based on a report by the Japanese Ministry of Health and Welfare,[13] coronary artery aneurysm (CAA) was defined as an actual internal diameter of ≥3 mm in a child aged <5 years or ≥4 mm in a child aged ≥5 years, the inner diameter of the segment being at least 1.5 times greater than that of an adjacent segment, or the luminal contour being irregular.

Ethical considerations

This study was conducted following the principles of the Declaration of Helsinki and the ethical guidelines issued by the Ministry of Health, Labour, and Welfare, Japan. The Ethics Committee of the Nagoya University Hospital approved the study (approval number: 2020-0452). Written informed consent was obtained from the guardians of the patients before participating in the study.

Statistical analyses

Nominal variables are shown as real numbers. Continuous data are expressed as the median and interquartile range (IQR) and first to third IQR. All statistical analyses were performed using EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan), a graphical user interface for R (The R Foundation for Statistical Computing, Vienna, Austria). It is a modified version of the R commander designed to add statistical functions frequently used in biostatistics.[14]

RESULTS

A total of 55 patients participated in the study. Table 1 shows the demographic data of patients. The study included 31 males (56.3%) and 24 females (43.7%). The median age was 1.6 (1.0–3.1) years, and the median body weight was 10.5 (9.1–13.1) kg. The median number of days of illness at initial treatment was 4 (4–5) days, and the median Kobayashi score was 3 (2–5) points. Three patients (5.4%) with the incomplete type were included. The first antibody measurement was performed at a median of 4th (3–5) days of illness, and the second was performed at a median of 12th (10–13) days of illness.

Table 1 Patient demographics

Variables	
Patients	55
Sex (Male/Female)	31/24
Body weight (kg)	10.5 (9.1–13.1)
Height (cm)	80.5 (73.0–93.4)
Age at diagnosis (years)	1.6 (1.0–3.1)
Incomplete type	3
Days of illness at initial treatment	4 (4–5)
Kobayashi score (points)	3 (2–5)
Days of illness at first antibody measurement	4 (3–5)
Days of illness at second antibody measurement	12 (10–13)

Data in median (interquartile range [IQR]) or number.

Table 2 Laboratory data of patients

Variables	Median (IQR), N=55
WBC ($\times 10^3/mm^3$)	13.7 (9.9–16.0)
% Neutrophils	65.0 (53.0–78.5)
Lymphocyte ($\times 10^3/mm^3$)	3.0 (1.9–4.9)
Platelet count ($\times 10^4/mm^3$)	33.1 (26.5–38.5)
AST (IU/L)	41 (30–64)
ALT (IU/L)	23 (14–80)
T-Bil (mg/dL)	0.5 (0.4–0.8)
ALB (g/dL)	3.8 (3.3–4.4)
Na (mmol/L)	134 (133–136)
CRP (mg/dL)	6.5 (2.5–10.7)

IQR: interquartile range
WBC: white blood cells
AST: aspartate aminotransferase
ALT: alanine transaminase
T-Bil: total bilirubin
ALB: albumin
Na: natrium
CRP: C-reactive protein

Table 2 shows the clinical laboratory data of the patients before initial treatment. Only two patients showed a low lymphocyte count (≤1000/mm^3), which makes up the diagnostic criteria for MIS-C by the Centers for Disease Control and Prevention (CDC).

Table 3 shows the results of anti-SARS-CoV-2 antibody measurements. In the measurements before KD treatment, regarding SARS-CoV-2 IgM, 48 cases (87.3%) had a COI <1. COI was slightly elevated between 1 and 2 in 7 cases (12.7%), and there were no positive cases with COI of ≥2. Regarding Elecsys Anti-SARS-CoV-2, only one case (1.8%) was positive, and this patient had a history of COVID-19 6 months before the onset of KD. In the measurements taken 10–14 days after the onset of KD, there were no positive cases of SARS-CoV-2 IgM. Elecsys Anti-SARS-CoV-2 was positive only in the same patient with a positive initial test result. Therefore, there was only one case in which the SARS-CoV-2 infection was confirmed before the onset of KD in this patient group.

Table 3 Antibody test results

Variables	<1.0	1.0–2.0	>2.0
Before the initial treatment (N=55)			
SARS-CoV-2 IgM (COI)	48	7	0
Elecsys Anti-SARS-CoV-2 (COI)	54	0	1
10–14th days of illness (N=55)			
SARS-CoV-2 IgM (COI)	50	5	0
Elecsys Anti-SARS-CoV-2 (COI)	54	0	1

COI: cutoff index

Table 4 treatment and prognosis

Variables	N=55
IVIG (%)	53 (96.3)
Initial combined use of steroid (%)	1 (1.8)
2nd line therapy (%)	12 (21.8)
2nd IVIG	12
mPSL pulse	3
Cyclosporine A	2
Infliximab	1
Heart failure or shock requiring innotrope	0 (0)
Gastrointestinal symptoms	0 (0)
No coronary artery complication (%)	53 (96.3)
CAA within 4 weeks after onset (%)	2 (3.6)
CAA at 4 weeks after onset (5)	1 (1.8)

IVIG: intravenous immunoglobulin
mPSL: methylprednisolone
CAA: coronary artery aneurysm

Table 4 shows the course of treatment and complications of KD. Fifty-three patients (96.3%) were treated with IVIG, and 12 (21.8%) received second-line therapy due to unresponsiveness to IVIG or relapse. Two patients presented with coronary aneurysms within 4 weeks of onset, one showed regression, and one had residual coronary aneurysms 4 weeks after onset. There were no cases of shock or heart failure requiring inotropes and gastrointestinal symptoms, which are included in the diagnostic criteria for MIS-C by CDC.

DISCUSSION

This study measured antibodies containing IgM and IgG against SARS-CoV-2 in 55 patients with KD at multiple hospitals during the COVID-19 pandemic. Most patients did not have elevated antibody levels. Studies investigating the association between SARS-CoV-2 infection and KD development by measuring SARS-CoV-2 antibody levels are limited. In this study, we investigated a multi-center patient population. We measured both SARS-CoV-2 IgM- and IgG-containing antibodies twice to examine the presence of SARS-CoV-2 infection in new patients with KD in detail. Our results suggest that SARS-CoV-2 infection before KD onset in this region was rare and that the association between KD onset and SARS-CoV-2 infection may be limited.

IgM and IgG antibodies against SARS-CoV-2 begin to increase during the week of COVID-19 symptoms onset and reach high levels by the third week.[13] In addition, elevated IgG levels persist beyond 7 weeks, in contrast to the almost complete disappearance of IgM by 7 weeks after onset.[15] Our study measured IgM and whole antibody levels, including IgG, at two points on the median 4th and 12th days of illness. Patients with negative IgM and IgG antibody levels at the two measurements were considered to have no antecedent SARS-CoV-2 infection for at least 7 weeks before KD onset. However, the sensitivity of SARS-CoV-2 IgM measurement reagent is 90.0% in the patients with confirmed diagnosis of SARS-CoV-2 by PCR or antigen tests.[10]

In addition, the sensitivity of Elecsys Anti-SARS-CoV-2 is 85.3% 7–13 days after a confirmed diagnosis of SARS-CoV-2 by PCR.[11] Therefore, especially in the case with infection just before the onset of KD, there is a possibility of a false negative result. Only one patient had elevated total antibody levels, including IgG, and a definite SARS-CoV-2 infection before the onset of KD. In this case, the patient had a history of COVID-19 6 months before, and the antibody elevation likely reflected this history. A long time has passed since the SARS-CoV-2 infection, and whether it was related to KD development in this patient was unknown.

Although the cause of KD remains unknown, it has been suggested that microbial infections such as viruses, bacteria, and fungi may trigger disease onset.[16] Regarding the association between other human coronaviruses before SARS-CoV-2 and KD, in 2005, Esper et al reported that a novel human coronavirus (HCoV-NL63) is associated with the development of KD.[17] However, contradictory results have been reported subsequently. Lehmann et al and Dominguez et al demonstrated HCoV-NL63 infection in 48% and 7.7% of patients with KD, respectively, using antibody measurements or polymerase chain reaction, although they did not differ from contemporaneous healthy controls.[18,19] Kondo et al measured serum IgG antibody levels against SARS-CoV-2 in KD patients at the time of admission around the same time as our study and similarly reported that few patients were positive.[20] Unlike the study by Kondo et al, we measured paired serum anti-SARS-CoV-2 both IgG and IgM antibodies and demonstrated that the antecedent infection, including the acute phase just before the onset of KD is quite rare. The IgG and IgM positivity rates in our study were lower than those reported in previous studies that examined the association between human coronaviruses and KD. Additionally, a decrease in the number of patients with KD has been reported since the COVID-19 pandemic in Japan.[7] These results suggest that the relationship between the prevalence of SARS-CoV-2 infection and the development of KD is limited.

Since the onset of the COVID-19 pandemic, there has been a series of reports of SARS-CoV-2 infections with symptoms resembling those of KD, reported as an MIS-C or pediatric inflammatory multisystem syndrome (PIMS) in the United States and Europe.[5,21] Symptoms overlap between MIS-C and KD, with reports suggesting that a quarter to half of the cases meet the diagnostic criteria for both KD and MIS-C.[22] In our study, none of the patients developed shock, heart failure, or gastrointestinal symptoms, the characteristics of MIS-C. Similarly, few patients had low lymphocyte and platelet counts, commonly observed in MIS-C.[23] In addition, the age range of patients in our study differed from that in MIS-C. In our study, the oldest patient was 6 years old, and many were infants. Since no cases in this study were suspected to overlap with MIS-C, it was possible to evaluate SARS-CoV-2 serum antibody levels in patients with pure KD. Rostad et al measured SARS-CoV-2 IgG levels in patients with MIS-C and KD and reported high IgG levels in all patients with MIS-C but not in those with KD.[24] Although our study findings are similar, we evaluated the antibody levels in more cases than the previous study did.

Our study had several limitations. First, this survey of four hospitals in one prefecture in Japan was not a population-based study. Therefore, the uneven distribution of SARS-CoV-2 epidemics and patients with KD may have influenced our results. However, each of the four participating hospitals was a core hospital in a different region that treats many patients with KD and COVID-19. Therefore, there was probably no bias in the background of patients with KD. Second, our study did not compare the antibody levels of patients with KD with that of healthy controls in the same geographic area during the same period. Therefore, it was impossible to precisely verify whether SARS-CoV-2 infection increased or decreased the risk of developing KD. Third, our study period is from April 2020 to July 2022, which includes only the epidemic period of the α, Δ, and ο strains of SARS-CoV-2. Whether similar results could be obtained with other variants after the study period requires further verification. Finally, this study was

conducted only in Japan and included only Asian patients. Racial differences in the risk of KD have been reported,[25] but our study could not determine whether the same results are observed in other races.

CONCLUSIONS

Anti-SARS-CoV-2 IgM and total antibody, including IgG, were measured in patients who developed KD during the COVID-19 pandemic. Except for one patient who had COVID-19 6 months before KD onset, no patient had elevated antibody levels, and in most patients, SARS-CoV-2 infection before the onset of KD was denied. Therefore, strong association between development of KD and SARS-CoV-2 infection was not suggested in our cohort.

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CONFLICT OF INTEREST

None.

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