

Prevalence of SARS-CoV-2 antibodies among university athletic club members: A cross-sectional survey

Yukihiro Mori^{1,2}, Mamoru Tanaka³, Hana Kozai³, Kiyoshi Hotta², Yuka Aoyama⁴, Yukihiro Shigeno^{5,6}, Makoto Aoike¹, Hatsumi Kawamura¹, Masato Tsurudome^{1,7}, Morihiro Ito^{1,7,*}

¹ Graduate School of Life and Health Sciences, Chubu University, Aichi, Japan;

² Center for Nursing Practicum Support, Chubu University, Aichi, Japan;

³ Department of Food and Nutritional Sciences, College of Bioscience and Biotechnology, Chubu University, Aichi, Japan;

⁴ Department of Clinical Engineering, College of Life and Health Sciences, Chubu University, Aichi, Japan;

⁵ Center for Emergency Medical Technician Practicum Support, Chubu University, Aichi, Japan;

⁶ The Fire Department Headquarters in Kasugai-City, Aichi, Japan;

⁷ Department of Biomedical Sciences, College of Life and Health Science, Chubu University, Aichi, Japan.

SUMMARY School-based coronavirus disease 2019 (COVID-19) testing is an important part of a comprehensive prevention strategy in public health. To assess the prevalence of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) antibodies in a university athletic club community with repeated occurrences of SARS-CoV-2 infections, we conducted a cross-sectional survey for asymptomatic antibody prevalence using a SARS-CoV-2 rapid antibody test kit. On January 26, 2021 we administered questionnaires to determine their history of contact with infected individuals and took blood samples from 129 undergraduates. The prevalence of SARS-CoV-2 antibodies among the subjects was 3.9%. Only 6.2% of the participants reported close contact with infected individuals. In this study, we clarified the prevalence of asymptomatic SARS-CoV-2 antibodies in university athletic clubs where SARS-CoV-2 infections had repeatedly occurred, which will be helpful in discussing how to identify and prevent the transmission of infections within university athletic club communities.

Keywords SARS-CoV-2 antibodies, university athletic club, COVID-19, asymptomatic antibody prevalence, SARS-CoV-2 rapid antibody test

1. Introduction

The appearance of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) in late 2019 led to the pandemic of coronavirus disease 2019 (COVID-19). As of June 2022, the pandemic has resulted in more than 530 million infected cases and over 6.3 million deaths worldwide. This has had the most serious global consequences on society, the economy, health systems, and human health since the 1918 influenza pandemic, which killed more than 50 million people.

In August 2020, a study in the United States reported a widespread outbreak of SARS-CoV-2 infection in a large university (1), and another study, in the next month, reported a rapid increase in SARS-CoV-2 infections in adults aged 18-22 years (2). With the increasing number of COVID-19 cases in college students, continuous preventive efforts, including strong testing regimes, were implemented to prevent

outbreaks on or near university campuses. The need for quarantine was emphasized to protect the wider community (3). School closures were imposed in many countries to prevent the spread of the disease to young people and their surroundings. In previous studies, school closures were associated with a temporary reduction in COVID-19 incidence and mortality (4).

COVID-19 mortality rates rise with age, with death being relatively rare among young adults (5). In general, the perception that young people have lower morbidity, severity, and mortality than older people can result in reduced awareness of infection control. When college students are infected with SARS-CoV-2, the potential for the rapid spread of the virus in the university environment is a significant concern, considering that students tend to have wider social networks than the general population.

Cluster infections of COVID-19 can occur in any population, including within families (6), communities

(7), and nursing homes (8). Educational institutions can be one such example (9). Therefore school-based COVID-19 testing is considered an important part of a comprehensive prevention strategy for identifying SARS-CoV-2 infections in schools and for maintaining face-to-face instruction and extra-curricular activities (10). Therefore, we conducted a cross-sectional screening survey of SARS-CoV-2 antibody prevalence in a university community. We also retrospectively investigated the contact history of the members of this community. This university community comprised students belonging to two athletic clubs. Many of the students at this university live in dormitories. We chose this community to study because of the elevated risk of outbreaks of not only COVID-19, but also other infectious diseases in these populations. The inclusion criterion was a lack of obvious symptoms of infection among the participants.

In this study, the primary objective was to assess the prevalence of asymptomatic SARS-CoV-2 antibodies among the students in university athletic clubs where COVID-19 had occurred repeatedly. The secondary objective was to observe the infection dynamics that can occur within university athletic clubs. Finally, to our knowledge, this is the first study to provide the prevalence of SARS-COV2 asymptomatic infections in athletic clubs of universities. We believe that these results could help identify and prevent the transmission of COVID-19 within university athletic club communities.

2. Materials and Methods

2.1. Participants

The flowchart of this study is shown in Figure 1. The participants were undergraduates belonging to either of the two athletic clubs at a private university in Japan. This university has an enrollment of approximately 12,000 students. The first confirmed COVID-19 infection in these athletic clubs was identified on July 14, 2020. Subsequently, 11 infections were confirmed from late November 2020 to mid-January 2021. This population was operationally defined in the present study as "the university athletic club with repeated occurrences of COVID-19 infection". A total of 129 students from these clubs without obvious symptoms of infection were included in the study; the COVID-19-infected students mentioned above were excluded. In Japan, vaccination with the COVID-19 mRNA vaccine began in early February 2021, therefore, none of the study populations had received the COVID-19 mRNA vaccine at the time we conducted our survey in January 2021.

2.2. SARS-CoV-2 antibody testing

SARS-CoV-2 antibody testing was performed on January 26, 2021, after the outbreak of infections in this club had ended. This period of blood collection was the first COVID-19 epidemics in Japan. We collected blood from the fingertips of the participants using safety lancets,

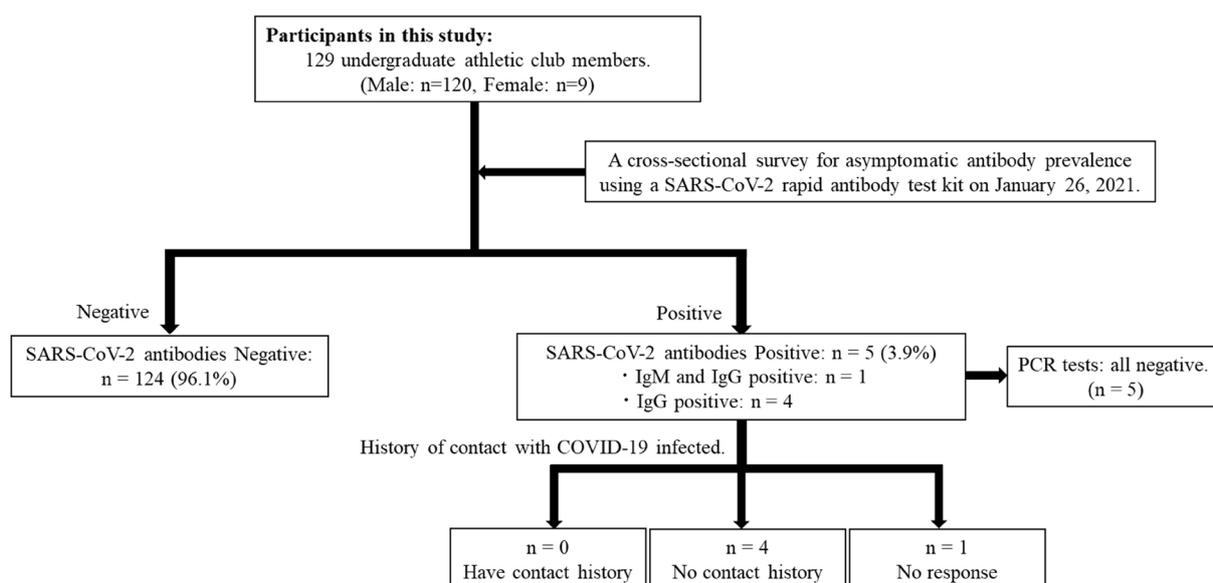


Figure 1. Study flow-chart. The first confirmed COVID-19 infection in this communities were identified on July 14, 2020. Subsequently, 11 infections were confirmed consecutively from late November 2020 to mid-January 2021. SARS-CoV-2 antibody testing was performed on January 26, 2021, after the outbreak of infections in this club had ended. We tested the participants ($n = 129$) for SARS-CoV-2 IgM and IgG antibodies using rapid antibody test kits. However, the selection criteria for the 129 subjects were those without obvious symptoms of infection, and COVID-19 patients were not included. Five blood samples (3.9%) tested positive for IgG antibodies, among which one sample was also positive for IgM antibodies. COVID-19, coronavirus disease 2019; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; IgM/G, Immunoglobulin M/G; PCR, polymerase chain reaction.

paying close attention to hygiene and infection control. The blood samples were tested using the SARS-CoV-2 Rapid Antibody Test RUO (Roche Diagnostics KK, Tokyo, Japan) by following the procedure on the package insert. This kit simultaneously detects SARS-CoV-2 IgG and IgM antibodies using immunochromatography. The procedure is as follows: the collected blood is placed into the sample drop hole of the test device, 3 drops (90 μ L) of buffer solution are added into the sample drop hole of the test device, and the results are read after 15 minutes. This test kit indicates that the patient may not be infected with SARS-CoV-2 if the line appears only on C (Control). The appearance of a line at G or M, together with C, indicates positivity for IgG or IgM antibodies specific for SARS-CoV-2, suggesting possible previous SARS-CoV-2 infection.

2.3. Reverse transcription-polymerase chain reaction (RT-PCR) testing

Saliva samples were collected from those who were positive for IgM and IgG antibodies in the SARS-CoV-2 antibody testing to perform real-time RT-PCR to check for SARS-CoV-2 infection. The Light Cycler 96 (Roche, Basel, Switzerland) and SARS-CoV-2 Direct Detection RT-qPCR Kit (TaKaRa, Siga, Japan) were used, and assessments were made by following the protocols. This kit uses the primer and probe sequence described in the "2019-Novel Coronavirus (2019-nCoV) Real-time RT-PCR Panel Primers and Probes" (Effective: Jan 24, 2020) published by the U.S. Centers for Disease Control and Prevention.

2.4. Survey

We surveyed the participants' attributes (age, sex), lifestyle habits (smoking and alcohol consumption), and medical history using a questionnaire. Participants were asked about contacts with individuals infected with SARS-CoV-2 in the last 6 months. The yes or no questions were: "Have any close relatives or anyone else around you been infected with COVID-19?" and for close contact, "I was in contact with a COVID-19 patient closer than 1 meter for 15 min or more without any infection prevention measures (mask, *etc.*)".

2.5. Statistical analysis

Microsoft Excel 2019 was used to aggregate the data collected and to calculate descriptive statistics. The data were primarily for exploratory purposes and are thus expressed as numbers and percentages.

2.6. Ethics

This study was approved by the ethics committee of the Chubu University (Approval No. 20200039). The study

was performed in accordance with the principles of the Declaration of Helsinki. The participants provided their written informed consent to participate in this study.

3. Results and Discussion

The participants included 120 men (93.0%) and 9 women (7.0%) with a mean age of 19.3 ± 1.0 years. Table 1 shows the participants' attributes, habits (smoking and alcohol consumption), and medical history. Six of the participants (4.7%) smoked and 21 (16.3%) drank alcohol. The most common medical complaint was asthma (17 participants, 13.2%), followed by pneumonia (5 participants, 3.9%), meningitis and heart disease (one each, 0.8%). Although we surveyed the participants' smoking and drinking habits and their medical history, there was no significant relationship between these factors and the prevalence of SARS-CoV-2 antibodies.

Results of the questionnaires revealed that 103 (80.6%) participants were aware of contacts infected with SARS CoV-2 and 26 (19.4%) were not. Among the 103 participants who answered "Yes", 94 (92.2%), did not have close contact with the infected person, eight (6.2%) had close contact and one (1.6%) did not respond. Participants were allowed to choose more than one option for relationship to infected contact, and the results showed that the vast majority (102, 99.0%) reported "a fellow club member" as the infected contact, one said "a friend outside the club," and one did not provide a response (Table 2).

We tested the participants ($n = 129$) for SARS-CoV-2 IgM and IgG antibodies. Five blood samples (3.9%) tested positive for IgG antibodies, of which one was also positive for IgM antibodies (Table 2). These antibody-positive samples were all from men (20.4 ± 0.8 years) and none of them reported any clinical symptoms associated with COVID-19. The PCR test results of

Table 1. Background characteristics of the study participants

Items	<i>n</i> (%)
Sex	
Male	120 (93.0)
Female	9 (7.0)
Smoking status	
Non-smoker	123 (95.3)
Former smoker	0 (0.0)
Current smoker	6 (4.7)
Alcohol	
Non-drinker	108 (83.7)
Drinker	21 (16.3)
Medical history*	
None	112 (86.8)
Asthma	17 (13.2)
Pneumonia	5 (3.9)
Meningitis	1 (0.8)
Heart disease	1 (0.8)

*Multiple responses were allowed.

saliva samples were negative in all five samples (100%) (Table 3).

All participants who tested positive for SARS-CoV-2 antibody (5 participants, 100%) were aware of the infected contacts, however four (80%) said that they had no history of close contact with the infected person, one (20%) did not respond. In addition, four (80%) said that they were fellow members of the same club for relationship to infected, one (20%) did not respond (Table 4).

We were inspired to conduct this study, because we wanted to determine the prevalence of asymptomatic COVID-19 infections and infection dynamics in one university athletic club with repeated COVID-19 infections. To the best of our knowledge, there have been no studies on the prevalence of asymptomatic COVID-19 infections in university athletic club communities where SARS-CoV-2 infections occurred. Our study revealed that the prevalence of asymptomatic COVID-19 infections was 3.9% in the university athletic club community where COVID-19 infections had repeatedly occurred. This result is consistent

with previous studies of university students and staff that reported SARS-CoV-2 antibody-positive rates of approximately 1-4% (11,12). However, a similar study conducted in Japan reported a rate of 1.23% (13). Our results were well above this value.

The first COVID-19 infection in this population was confirmed in mid-July 2020. Subsequently, 11 infections were confirmed from late November 2020 to mid-January 2021. To retrospectively track the contact history of this community, we conducted a questionnaire survey along with SARS-CoV-2 antibody testing of serum samples. The results showed that about 80% of the participants were aware of an infected person nearby, and only about 6% had close contact with the infected individual. Because of the lack of a comparison group, it is unclear whether this contact rate is higher than that in other communities.

In Japan, priority vaccination with the COVID-19 mRNA vaccine began in early February 2021 for approximately 4.8 million healthcare workers, therefore, none of the study populations had received the COVID-19 mRNA vaccine at the time we conducted our survey in January 2021. Therefore, the data obtained here provide valuable information on the prevalence of asymptomatic SARS-CoV-2 antibodies among members of athletic clubs prior to the widespread use of the COVID-19 mRNA vaccine.

In addition, since the club activities continued during

Table 2. SARS-CoV-2 antibody test results, presence or absence of infected individuals in the vicinity; contact history and relationship with infected individuals

Items	n (%)
SARS-CoV-2 antibodies	
IgM Negative	128 (99.2)
Positive	1 (0.8)
IgG Negative	124 (96.1)
Positive	5 (3.9)
Awareness of infected individuals in the vicinity	
Yes	103 (80.6)
No	26 (19.4)
Contact history [†]	
Yes	8 (6.2)
No	94 (92.2)
No response	1 (1.6)
Relationship ^{††}	
Fellow club member	102 (99.0)
Friend outside club	1 (1.0)
No response	1 (1.0)

SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; IgM/G, Immunoglobulin M/G. [†]Contact history and Relationships, n = 103.

^{††}The relationship question allowed for multiple responses.

Table 4. Presence/absence of infected individuals in the vicinity of SARS-CoV-2 antibody-positive individuals, contact history, and relationship

Items	n (%)
Awareness of infected individuals in the vicinity	
Yes	5 (100)
No	0 (0)
Contact history	
Yes	0 (0)
No	4 (80.0)
No response	1 (20.0)
Relationship	
Fellow club member	4 (80.0)
Friend outside club	0 (0)
No response	1 (20.0)

SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

Table 3. Attributes of SARS-CoV-2 antibody-positive individuals, detection of SARS-CoV-2 IgM and IgG antibodies

Case No.	Age	Sex	Medical history	SARS-CoV-2 PCR	COVID-19 Symptoms	SARS-CoV-2 Antibodies	
						IgM	IgG
1	21	Male	Asthma	Negative	None	+	+
2	21	Male	-	Negative	None	-	+
3	20	Male	-	Negative	None	-	+
4	20	Male	-	Negative	None	-	+
5	19	Male	Asthma, pneumonia	Negative	None	-	+

+, positive; -, negative; COVID-19, coronavirus disease 2019; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; PCR, polymerase chain reaction; IgM/G, Immunoglobulin M/G.

the COVID-19 pandemic, university administrators and coaches must implement preventive measures, such as encouraging appropriate social distancing, conducting daily health checks, and measuring body temperature (14). Furthermore, many members of these athletic clubs live in student dormitories which are places where crowded situations can occur easily. The risk of spreading COVID-19 infections in shared living environments, such as dormitories, has long been pointed out (15).

Club activities are an essential element of university life, the suspension of club activities during the COVID-19 pandemic can affect the members' progress and be a source of serious anxiety. Moreover, it has been pointed out that exercising during a pandemic has a positive effect on mental health (16). Therefore, university administrators and club coaches should try to maintain opportunities for exercise in order to maintain the students' physical and mental health. As such, strategies to prevent the spread of COVID-19 in club communities, in addition to robust infection control measures, must involve the creation of an environment that enables monitoring of SARS-CoV-2 antibody prevalence through continuous testing.

This study had several limitations. 1) The specificity declared by the manufacturer of the SARS-CoV-2 rapid antibody test kit used in this study was 98.65%, with a sensitivity of 99.03% for cases after 14 days of illness. However, SARS-CoV-2 antibody tests can produce contradictory results depending on the assay used (17,18). 2) Differences in the positive rates of SARS-CoV-2 antibodies can arise due to factors, such as country, region, community, race (19), socioeconomic environment (20), and cultural background (21). 3) Due to the limited number of samples we were able to collect, the sample size for this study was not large, so caution should be exercised when generalizing the results. In addition, the male-female ratio in this population was 40:3 and the age group was 19.3 ± 1.0 years, which makes our results difficult to compare with those of previous studies (22), which have observed age- and sex-based differences in SARS-CoV-2 antibody responses. A variety of other potential biases were not addressed, such as participation in social activities outside of the club activities and district of residence. 4) In the questionnaire survey, it should be noted that the participants' knowledge or awareness of infected people nearby and their contact history were based on self-reports. Regarding the operational definition of contact history, in particular, there is limited evidence supporting its accuracy. In addition, although a low percentage of participants had a history of contact with infected people, the results of this study do not necessarily mean that it was more likely that exposure came from outside the community. Multiple previous studies that investigated contact history through questionnaires and interviews have also

designated operational definition (23,24). In discussing the relationship between SARS-CoV-2 antibody prevalence within communities, it is important to carefully consider how to devise tracking methods, set operational definitions, and consider the subjective bias of the participants with regard to the contact history data obtained from questionnaires and interviews. 5) Because this was a cross-sectional study, we did not follow up on the prevalence of SARS-CoV-2 antibodies in the participants' community. 6) Finally, this study represents provisional results as of January 2021, and results may change due to future epidemics, changes in the state of control, and the impact of new variants.

In summary, the present study provides novel and unique data on the prevalence of asymptomatic COVID-19 infections within a community of university athletic clubs where infections had repeatedly occurred. This is a strength of this study. These findings could be useful for comparative data for future studies involving athletic club communities.

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- *Address correspondence to:
Morihito Ito, Department of Biomedical Sciences, College of Life and Health Sciences, Chubu University, 1200 Matsumoto-cho, Kasugai, Aichi 487-8501, Japan.
E-mail: m-ito@isc.chubu.ac.jp
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