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Effect of COVID-19 vaccination on viral clearance and antibody production in older patients with SARS-CoV-2 infection

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ABSTRACT

Whether coronavirus disease 2019 (COVID-19) vaccination promotes viral clearance in older patients has not been reported. We performed a retrospective review of patients hospitalized with COVID-19. This study included 24 patients with COVID-19 admitted to Hiroshima City Funairi Citizens Hospital between June 1 and July 10, 2021. Nine patients who were vaccinated (median age: 72 years) were compared with 15 patients who were not vaccinated (median age: 70 years). Viral clearance was confirmed by SARS-CoV-2 reverse transcription polymerase chain reaction (RT-PCR). Antibody titers were measured to assess vaccination efficacy. The vaccinated group had a higher negative conversion rate than that in the non-vaccinated group on RT-PCR testing before discharge (83% vs. 36%, $P = 0.064$). Antibody titers on admission and 10 ± 2 days after onset were significantly higher in the vaccinated group than those in the non-vaccinated group (35 vs. 0 binding antibody units (BAU)/mL, $P = 0.012$; and 114 vs. 7 BAU/mL, $P = 0.032$, respectively). Stimulating antibody production by vaccination may promote faster viral clearance in older patients who develop COVID-19.

Key words: *COVID-19*, *Older patients*, *Vaccine*, *Viral clearance*

INTRODUCTION

The first confirmed case of coronavirus disease 2019 (COVID-19) was identified in Wuhan, China, in December 2019, and the disease subsequently became a pandemic. To control the COVID-19 pandemic, messenger RNA (mRNA)-based vaccines have been developed by Moderna (mRNA-1273) and Pfizer-BioNTech (BNT162b2), both of which have been found to have 95% effectivity at preventing severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection[1,14]. These vaccines were designed to produce antibodies targeting the SARS-CoV-2 spike protein, inhibiting entry of the virus into host cells[6]. A recent study showed that even a single dose of vaccine significantly reduced symptom duration and time to viral clearance in healthcare workers with SARS-CoV-2 infection, with an average age of approximately 40 years[3]. However, it has not been reported whether a single dose of the vaccine is effective at promoting viral clearance in older patients.

Previous studies have reported that a longer duration was required for viral clearance in older patients with COVID-19 than in younger patients[17,18], and older age, male sex, delayed hospital admission after illness onset, and hypertension have been identified as risk factors for delayed viral clearance[2,19,20]. Thus, it is important to determine whether vaccines are effective in older patients.

In Japan, older people are given priority for vaccination because they are more prone to developing severe disease. With the increase in vaccination rates in older people, the proportion of older patients with COVID-19 has declined remarkably[11]. According to this observation, vaccination may not only prevent infection, but may also be effective in promoting early viral clearance, even in older people.

This retrospective study aimed to determine whether sufficient antibody production occurs in older people and whether vaccination promotes rapid viral clearance in older patients who develop COVID-19.

PATIENTS AND METHODS

Patients

Ninety-three patients with COVID-19 were admitted to Hiroshima City Funairi Citizens Hospital between

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Table 1 Characteristics of the vaccinated patients (N = 9)

Characteristic	Value
Age (years), median (range)	72 (50–90)
Male/female	8/1
Comorbidities	
Hypertension	2
Diabetes mellitus	1
Cerebral infarction	2
Rheumatoid arthritis	1
After one dose of vaccine	7 patients
Days from the last vaccination to positive RT-PCR, median (range)	10 (5–18)
After two doses of vaccine	2 patients
Days from the last vaccination to positive RT-PCR, median (range)	8.5 (5–12)
Adverse event of vaccine	
Fever and malaise	1
Inoculation site pain	3
None	5

RT-PCR, reverse transcription polymerase chain reaction.

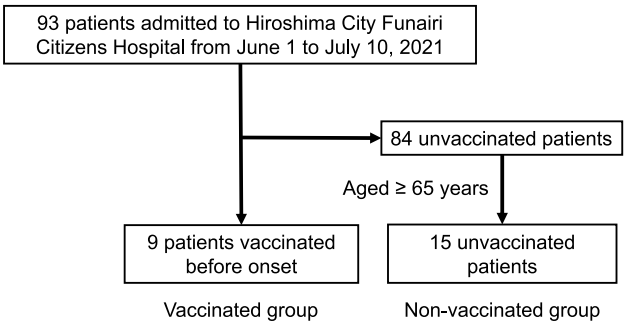

Figure 1 Flowchart of patient enrollment.

June 1 and July 10, 2021. All patients were diagnosed with COVID-19 based on positive SARS-CoV-2 real-time reverse transcription polymerase chain reaction (RT-PCR) results.

Of these 93 patients, 9 patients who had received vaccination before admission were enrolled in the vaccinated group (median age: 72 years, range: 50–90 years). Of the remaining 84 patients, all of whom had not been vaccinated, 15 (aged ≥ 65 years) were selected and enrolled in the non-vaccinated group (median age: 70 years, range: 65–97 years) to make the groups similar in terms of median patient age (Figure 1).

The procedures followed were in accordance with the Declaration of Helsinki, and the study was approved by the Institutional Review Board of Hiroshima City Funairi Citizens Hospital (approval no. 2021004).

Measurement of viral clearance (negative conversion)

Viral clearance was confirmed by SARS-CoV-2 RT-PCR using nasopharyngeal or salivary samples. RT-PCR tests were performed with patient consent using Takara SARS-CoV-2 Direct PCR detection kit (TaKaRa Bio Inc., Shiga, Japan) and LightCycler® 480 System II (Roche Diagnostics, Basel, Switzerland). RT-PCR results were regarded as negative if the cycle threshold (Ct) value was > 40 or not detected.

Antibody titer measurement

Serum anti-SARS-CoV-2 IgG antibody titers against spike protein S1 were measured in most patients during hospitalization. The VITROS® Anti-SARS-CoV-2 S1 Quant IgG kit (Ortho Clinical Diagnostics, USA) was used to determine the antibody titer, and binding antibody units (BAU) were used to indicate the titer according to the World Health Organization international standard (values ≥ 17.8 considered positive; upper limit 4,000).

Statistical analysis

An unpaired *t*-test or Wilcoxon rank-sum test was performed to compare the continuous variables of the two groups, as appropriate, and chi-square tests were used to compare categorical variables. Unless otherwise stated, all results were presented as the median (range). Statistical significance was set at $P < 0.05$. All statistical analyses were performed using JMP Pro 14.2.0® (SAS Institute Inc., Cary, NC, USA) statistical software.

RESULTS

Patient characteristics

The characteristics of the vaccinated group are shown in Table 1. The vaccinated group included eight men and one woman. Seven patients (six men and one woman) received one dose of the vaccine, and two patients (men) received two doses. The days from the last vaccination date to the date of positive RT-PCR were 10 days and 8.5 days in patients who had received one and two doses of vaccine, respectively. Comorbidities included hypertension, diabetes mellitus, cerebral infarction, and rheumatoid arthritis (treated using methotrexate [MTX]). As adverse events of the vaccination, three patients (all men) had experienced inoculation site pain, and one patient (man) had experienced fever and malaise.

The two groups (vaccinated and non-vaccinated) are compared in Table 2. The proportion of men and serum C-reactive protein (CRP) levels on admission were signif-

Table 2 Comparison of clinical features between the vaccinated and non-vaccinated groups

Patient characteristics	Non-vaccinated group n = 15	Vaccinated group n = 9	*P*-value
Age, years	70 (65–97)	72 (50–90)	0.455
Sex			
Male	7 (47%)	8 (89%)	0.039
Female	8 (53%)	1 (11%)	
Smoking habit	8 (53%)	6 (67%)	0.521
BMI, kg/m^2	23.2 (16.9–33.6)	23.2 (18.4–28.7)	0.912
Comorbidities			
Hypertension	4 (27%)	2 (22%)	0.808
Diabetes mellitus	3 (20%)	1 (11%)	0.572
Chronic kidney disease	1 (7%)	1 (11%)	0.703
Respiratory disease	2 (13%)	0 (0%)	0.253
SpO_2 at admission < 93%	7 (47%)	3 (33%)	0.521
Symptoms			
Fever	15 (100%)	8 (89%)	0.187
Cough	9 (60%)	7 (78%)	0.371
Fatigue	8 (53%)	5 (56%)	0.916
Dyspnea	3 (20%)	2 (22%)	0.897
Duration from illness onset to admission, days	6 (1–10)	3 (0–8)	0.051
Pneumonia on chest CT on admission	15 (100%)	9 (100%)	> 0.99
Laboratory values			
WBC (10^3/μL)	4.6 (3.2–6.5)	4.3 (3.0–10.6)	0.633
Neutrophils (10^3/μL)	3.2 (2.1–4.7)	3.2 (1.5–9.3)	0.953
Lymphocytes (10^3/μL)	1.0 (0.52–1.8)	0.77 (0.37–1.6)	0.240
CRP (mg/dL)	2.47 (0.05–9.41)	5.27 (0.18–14.6)	0.044
IL-6 (pg/mL)	35.5 (15.6–101)	57.8 (4.2–151)	0.191
Ferritin (ng/mL)	422 (21–1,803)	204 (92–2,143)	0.905
Treatment			
Remdesivir	13 (87%)	8 (89%)	0.873
Steroid	11 (73%)	6 (67%)	0.728
Baricitinib	2 (13%)	2 (22%)	0.572
Oxygen support	8 (53%)	4 (44%)	0.673
Length of hospital stay, days	9 (5–28)	10 (6–12)	0.185
Clinical outcome			
Discharged	13 (87%)	8 (89%)	0.873
Transferred to a general ward	2 (13%)	1 (11%)	0.873
Death	0 (0%)	0 (0%)	> 0.99

The results were reported as frequency (%) or median (range). BMI, body mass index; CRP, C-reactive protein; CT, computed tomography; IL-6, interleukin 6; SpO_2, oxygen saturation; WBC, white blood cells.

icantly higher in the vaccinated group than that in the non-vaccinated group (men: 8/9 [89%] vs. 7/15 [47%], *P* = 0.039; CRP: 5.27 vs. 2.47 mg/dL, *P* = 0.044). However, there were no statistically significant differences between the two groups in terms of their other background characteristics, treatment, length of hospital stay, or clinical outcomes (Table 2).

Negative conversion rate on RT-PCR testing before discharge

Six of 9 patients in the vaccinated group (67%) and 11 of 15 patients in the non-vaccinated group (73%) underwent RT-PCR testing before discharge (Table 3). Although the duration from onset to RT-PCR before discharge was a few days longer in the non-vaccinated group than in the vaccinated group (14 days and 11.5 days, respectively), this difference was not statistically significant (*P* = 0.158). The negative conversion rate on RT-PCR before discharge was higher in the vaccinated group (83%) than in the non-vaccinated group (36%), but this difference was not statistically significant (*P* = 0.064) (Table 3).

Figure 2 shows the changes in Ct values over time since illness onset in the vaccinated group (N = 6) and the non-vaccinated group (N = 11). Patients in the vaccinated group tended to show a more rapid increase in Ct values than those in the non-vaccinated group.

Anti-SARS-CoV-2 IgG antibody titers

On admission, anti-SARS-CoV-2 IgG antibody titers were positive in six of nine patients in the vaccinated group and in only three of 15 patients in the non-vaccinated group. Table 4 shows the intra-individual changes in the antibody titers from admission day and before discharge in the vaccinated group. The antibody test on admission was negative in patients admitted

Table 3 Comparison of RT-PCR results performed before discharge between the vaccinated and non-vaccinated groups

	Non-vaccinated group n = 15	Vaccinated group n = 9	*P*-value
RT-PCR performed before discharge	n = 11 (73%)	n = 6 (67%)	
Duration from onset to RT-PCR before discharge, days	14 (10–19)	11.5 (9–14)	0.158
Negative RT-PCR before discharge	4 (36%)	5 (83%)	0.064

The results were reported as frequency (%) or median (range). RT-PCR, reverse-transcription polymerase chain reaction.

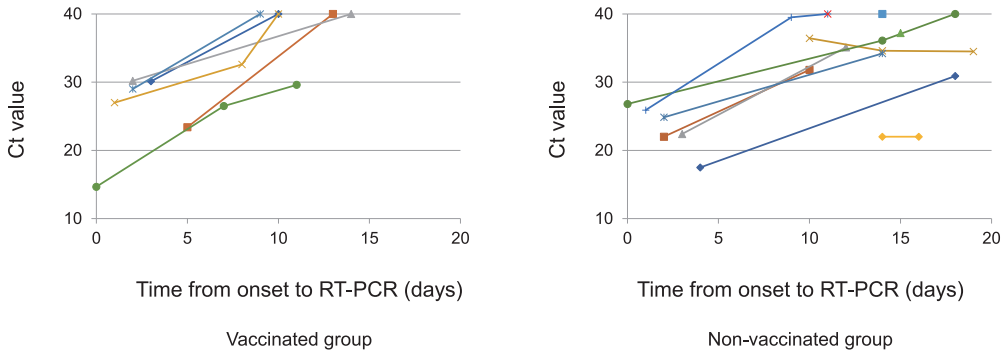

Figure 2 Changes in the cycle threshold (Ct) values in reverse transcription polymerase chain reaction (RT-PCR). A. Changes in Ct values in the vaccinated group. B. Changes in Ct values in the non-vaccinated group. Ct: cycle threshold, RT-PCR: reverse transcription polymerase chain reaction.

Table 4 Changes in the anti-SARS-CoV-2 IgG antibody titers in vaccinated patients

Patient no.	Number of vaccine doses	Days from first vaccination to hospitalization	Days from the onset to hospitalization	Antibody titer during hospitalization			RT-PCR before discharge
				Days 0–1 (BAU/mL)	Days 2–4 (BAU/mL)	Days 5–8 (BAU/mL)	
1	2	37	1	35	60	88	+
2	2	26	1	334	3,580	4,000	–
3	1	19	2	33	44	789	N/A
4	1	18	3	175	900	3190	–
5	1	18	8	74	82	N/A	N/A
6	1	13	8	307	594	1720	–
7	1	8	7	0	5	162	–
8	1	7	3	1	4	93	–
9	1	6	0	0	0	114	N/A

BAU, binding antibody units; N/A, not available; RT-PCR, reverse transcription polymerase chain reaction; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2.

within 8 days after the first vaccination (Patients no. 7, 8, and 9) and positive in those admitted 13 days after the first vaccination (Patients no. 1, 2, 3, 4, 5, and 6). One patient (Patient no. 1) in the vaccinated group sustained a positive RT-PCR result before discharge. He had rheumatoid arthritis that was treated with MTX. Despite receiving two doses of the vaccine and having the longest duration from the first vaccination to hospitalization, the patient showed the lowest antibody titer on days 5–8 of hospitalization.

Figure 3 shows a comparison of the anti-SARS-CoV-2 IgG antibody titers between the two groups. On the day of admission, the median values of anti-SARS-CoV-2 IgG antibody titers were significantly higher in the vaccinated group (median: 35 BAU/mL; range: 0–334 BAU/mL) than in the non-vaccinated group (median: 0 BAU/mL; range: 0–186 BAU/mL; P = 0.012). Anti-SARS-CoV-2 IgG antibody titers were retested 10 ± 2 days from illness onset in 9 of the total patients in the vaccinated group and 13 of 15 patients in the non-vaccinated group. The titers were significantly higher in the vaccinated group (median: 114 BAU/mL, range: 35–4,000 BAU/mL) than in the non-vaccinated group (median: 7 BAU/mL, range: 0–1,080 BAU/mL) on 10 ± 2 days from the onset (P = 0.032) (Figure 3).

DISCUSSION

To the best of our knowledge, this is the first study to report the effects of vaccination on viral clearance and the production of anti-SARS-CoV-2 IgG antibodies in older patients with COVID-19.

We obtained two notable results. First, older patients with COVID-19 who had received at least one dose of the vaccine showed a higher negative conversion rate on RT-PCR performed before discharge than patients

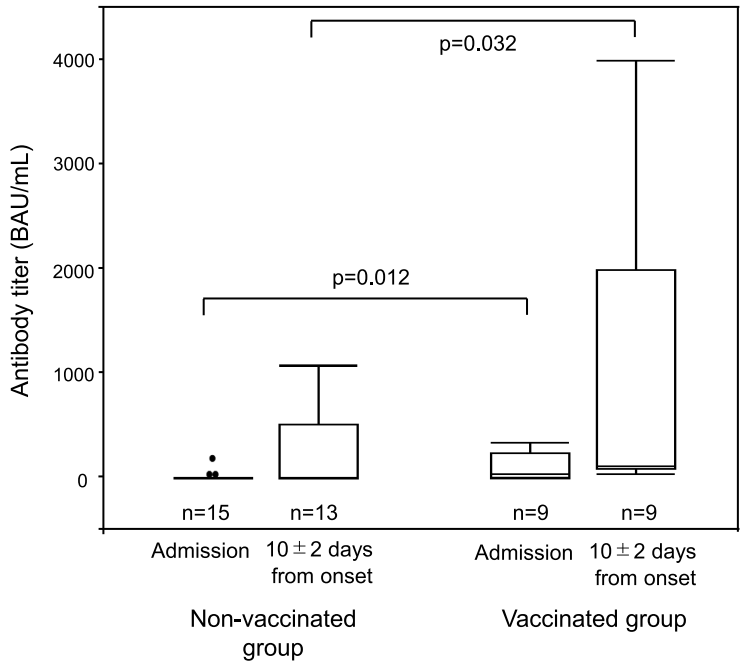

Figure 3 Comparison of the antibody titers in the non-vaccinated group (left) and the vaccinated group (right) on admission and 10 ± 2 days from onset. BAU: binding antibody units.

who were not vaccinated. This result suggests that vaccination promotes viral clearance in older patients with COVID-19. Second, the vaccinated group showed a significantly greater increase in anti-SARS-CoV-2 IgG antibody titers during hospitalization than the non-vaccinated group.

The relationship between humoral immunity and viral clearance is well known, and there are two mechanisms whereby SARS-CoV-2 antibodies promote viral clearance:1) the antibodies bind to the SARS-CoV-2 spike protein to neutralize infection by directly blocking the viral replication cycle and 2) the antibodies trigger complement activation and antibody-dependent cellular cytotoxicity[5,6,16].

Several studies have focused on the clinical relationship between antibodies and SARS-CoV-2 clearance[8,9,15]. Inpatients with COVID-19 who were antibody-positive had significantly higher Ct values on admission than antibody-negative patients ($p < 0.0001$), suggesting that antibodies contributed to the reduction in the patients' viral load[8]. In another study, there was also a correlation between high levels of anti-SARS-CoV-2 IgG titers and low viral load observed in vaccinated healthcare workers[15]. In addition, there is a positive correlation between time to antibody reaction and time to viral clearance; earlier antibody production is associated with more rapid viral clearance[9]. Our results showed that the increase in antibody titers was significantly higher in the vaccinated group than in the non-vaccinated group, suggesting that the increase in antibody titer in the vaccinated group was driven mainly by vaccination. As the negative conversion rate in patients with COVID-19 was higher in older patients in the vaccinated group than in the non-vaccinated group, it appears that the antibody-producing activity enhanced by vaccination may play a clinically significant role in promoting viral clearance.

The negative effect of MTX on the humoral response to vaccination[4,7,10] is a possible reason why one patient in the vaccinated group, who was being treated for rheumatoid arthritis with MTX, had a positive RT-PCR result before discharge. This patient had a relatively low increase in antibody titers, although he had received two doses of the vaccine (Patient no. 1 in Table 4). Thus, clinicians must be aware that vaccination may be less effective at stimulating antibody production and promoting viral clearance in immunosuppressed patients.

This study has several limitations. First, this was a single-center study with a small sample size; the study participants were selected only among hospitalized patients, and patients with mild symptoms were not included. While vaccination is known to prevent aggravation of the COVID-19 disease state[13], it would be interesting to determine whether the effect of vaccination on viral clearance and antibody production differs according to disease severity among vaccinated patients.

Second, we were unable to evaluate whether the effect of vaccination on viral clearance varied according to the variant of the virus. During the study period, the majority of SARS-CoV-2 infections were caused by the alpha variant[12]; delta and omicron variants had not yet emerged. Our findings may not be applicable to cases in which these variants have avoided the antibodies produced after vaccination because of mutations in SARS-CoV-2 spike proteins.

In conclusion, in older patients with COVID-19, vaccination before SARS-CoV-2 infection may promote more rapid viral clearance because of the effect of vaccination on the humoral immune response. This study demonstrates that even a single dose of vaccine has a beneficial effect on antibody production and viral elimination. Further research is warranted to clarify the clinical significance of vaccination for viral clearance.

Conflicts of Interest

The authors declare no conflict of interest.

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