Received: 7 July 2022 | Revised: 18 September 2022 | Accepted: 20 September 2022

DOI: 10.1111/cas.15603

ORIGINAL ARTICLE

Cancer Science WILEY

Attenuated humoral response against SARS-CoV-2 mRNA vaccination in allogeneic stem cell transplantation recipients

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Abstract

Antibody persistence several months after severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) mRNA vaccination in allogeneic stem cell transplantation recipients remains largely unknown. We sequentially evaluated the humoral response to two doses of mRNA vaccines in 128 adult recipients and identified the risk factors involved in a poor response. The median interval between stem cell transplantation and vaccination was 2.7 years. The SARS-CoV-2 S1 Ab became positive after the second vaccination dose in 87.6% of the recipients, and the median titer was 1235.4 arbitrary units (AU)/ml. In patients on corticosteroid treatment, the corticosteroid dose inversely correlated with Ab titer. Multivariate analysis identified risk factors for poor peak response such as an interval from stem cell transplantation ≤1 year, history of clinically significant CMV infection, and use of >5 mg/day prednisolone at vaccination. Six months after vaccination, the median titer decreased to 185.15 AU/ml, and use of >5 mg/day prednisolone at vaccination was significantly associated with a poor response. These results indicate that early vaccination after stem cell transplantation (<12 months) and CMV infection are risk factors for poor peak response, while steroid use is important for a peak as well as a persistent response. In conclusion, although humoral response is observed in many stem cell transplantation recipients after two doses of vaccination, Ab titers diminish with time, and factors associated with persistence and a peak immunity should be considered separately.

KEYWORDS
allogeneic stem cell transplantation, COVID-19, humoral immunity, mRNA vaccine, SARS-CoV-2

Abbreviations: ATG, antithymocyte globulin; AU, arbitrary unit; cGVHD, chronic GVHD; CI, confidence interval; COVID-19, coronavirus disease 19; csCMVi, clinically significant CMV infection; GMT, geometric means titer; GVHD, graft-versus-host disease; OR, odds ratio; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; SCT, stem cell transplantation; TKI, tyrosine kinase inhibitor.

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1 | INTRODUCTION

Allogeneic hematopoietic SCT recipients are immunocompromised and are at a high risk of developing critical COVID-19, which is caused by SARS-CoV-2.[1–3] Excellent efficacy and safety of two doses of mRNA vaccines in healthy individuals have been reported,[4,5] although chronological deterioration of immune responses in vaccinated people remains a matter of investigation.[6,7] Regarding the risk of aggravation of COVID-19[1,2,8] and the insufficient response after the second vaccination due to long-lasting immunodeficiency in and administration of immunosuppressive agents to SCT recipients,[9–11] pertinent evaluation of the peak response as well as persistence of vaccine response is essential for optimal patient care. However, previous studies have mainly focused on the peak titer or humoral response at a single time point, and the analysis of immune longevity, including determinants, is limited.[12] In addition, some studies have reported an association between immunosuppressive agent use and a poor vaccine response,[10,13] although the details remain unclear. The main objective of this study was to evaluate the humoral response to SARS-CoV-2 mRNA vaccines in 128 SCT recipients and the secondary objective was to identify the risk factors associated with poor peak and/or persistence of vaccine response in this population.

2 | MATERIALS AND METHODS

2.1 | Patients

In this single-center prospective observational study, patients older than 18 years of age who underwent allogeneic SCT at our hospital, received two doses of the SARS-CoV-2 mRNA vaccines (BNT162b2 or mRNA-1273), and agreed to participate in the study, were enrolled. The first dose of the vaccination was administered between June 2021 and November 2021, and only the BNT162b2 and mRNA-1273 vaccine types were available in Japan during the study period. All SCT recipients who visited our hospital as outpatients were eligible for inclusion. However, the following were excluded: patients who were intravenous immunoglobulin preparation-dependent during vaccination, did not receive a second vaccination dose within 42 days after the first dose, and did not visit the hospital for sample collection. Patients undergoing prophylactic administration of chemotherapy were eligible for inclusion, but patients with hematological relapse were excluded. In general, the interval between the first and second doses of the vaccine was 3 weeks for BNT162b2 and 4 weeks for mRNA-1273. Serum samples were serially collected before vaccination, 2 weeks ± 7 days after the first vaccination dose, 5 or 6 weeks after the first vaccination dose (i.e., 2 weeks after the second vaccination dose), 3 months ± 1 month after the first vaccination dose, and 6 months ± 1 month after the first vaccination dose. The detailed transplantation procedures have been previously described.[14] Clinically significant CMV infection was defined as the initiation of preemptive anti-CMV therapy or onset of CMV end-organ disease. The severity of COVID-19 was classified according to WHO recommendations.[15]

This study was carried out in accordance with the Declaration of Helsinki and approved by the Ethics Committee of Tokyo Metropolitan Komagome Hospital (approval number 2716). Written informed consent to participate in the study was obtained from all patients.

2.2 | Evaluation of anti-SARS-CoV-2 Ab titer

The concentration of anti-SARS-CoV-2 IgG was measured in serum samples at each time point using an iFlash 3000 chemiluminescence immunoassay analyzer (Shenzhen YHLO Biotech) with an iFlash–SARS-CoV-2 IgG kit and iFlash–SARS-CoV-2 IgG-S1 kit, as previously described.[16] The iFlash-SARS-CoV-2 IgG kit primarily detects anti-nucleocapsid Abs;[16] therefore, patients with positive SARS-CoV-2 IgG results were considered to have a previous history of COVID-19 and were excluded from analyses. The iFlash–SARS-CoV-2 IgG-S1 kit detects IgG specific to the S1 subunit of the spike (S) protein. According to the manufacturer's instructions, samples with values ≥10 AU/ml were considered positive; in addition, a value >500 AU/ml was defined as a high titer. The following conversion formula applied: YHLO IgG-S: AU/ml × 1 = BAU/ml.

2.3 | Statistics

Differences in numerical and categorical variables were compared using the *t*-test and Fisher's exact test, respectively. Correlations from Fisher's exact test were plotted with transition colors (red [positive] and blue [negative] correlations), with the circle size indicating the *p* value. Spearman's rank correlation coefficient was used to determine the correlations between the two numerical variables. Logistic regression analysis was carried out for multivariate analysis. Factors with $p < 0.05$ in univariate analyses were included in the multivariate analysis. The cumulative incidence of COVID-19 was evaluated using Gray's method, considering relapse and death as competing risks. The level of statistical significance was set at $p < 0.05$. Statistical analyses were undertaken using R version 4.0.3.

3 | RESULTS

3.1 | Patient characteristics

One hundred and forty-two patients were included in this study. Of these, 14 were excluded from the analysis for the following reasons: history of COVID-19 before or within 14 days after vaccination ($n = 7$), unavailability of records showing that the second vaccination dose was given within 42 days after the first dose ($n = 2$), occurrence of severe hypogammaglobulinemia after inclusion requiring regular

treatment with intravenous immunoglobulin (n = 2), rejection of vaccination after inclusion (n = 2), and disease relapse after inclusion (n = 1). The number of serum samples at each time point was 120 before vaccination; 29, 2 weeks after the first vaccination dose; 128, 2 weeks ± 7 days after the second vaccination dose (5–6 weeks after the first dose); 119, 3 months after the first vaccination dose; and 67, 6 months after the first vaccination dose. We noted that the number of collected samples, particularly those after the first vaccination dose, was small because the epidemic and the unstable medical systems prevented frequent hospital visits. The median interval (with first and third quartiles) between the first and second vaccination doses was 21 days (21, 22) for BNT162b2 and 28 days (28–31) for mRNA-1273. The median interval (with first and third quartiles) from the first vaccination dose to each sampling was 14.5 (13.0–17.0) days after the first vaccination, 38.0 (35.0–41.0) approximately 5 or 6 weeks after the first vaccination dose, 96.0 (89.0–103.0) at 3 months after vaccination, 180.0 (170.5–187.5) at 6 months after the first vaccination dose, and the median interval from the second vaccination dose to sampling after the second vaccination was 15.0 (12.0–19.0) days.

Details of patient characteristics are shown in Table 1. The median age at vaccination was 54.0 years (range, 22–76 years), and the median interval from SCT to the first vaccination dose was 2.7 years (range, 0.3–31.3 years). Only one patient had a vaccine at less than 6 months after SCT. The type of vaccine used was mainly BNT162b2 (83.6%). Rituximab was given before SCT to five patients; the interval between rituximab initiation and vaccination was 7 months for one patient and more than 4 years for four patients. Most haploidentical SCT recipients underwent ATG-based SCT[17] rather than post-transplant cyclophosphamide treatment. Thirty-one patients were administered ATG for GVHD prophylaxis. Forty-seven patients had a history of grade II–IV acute GVHD, and moderate/severe cGVHD was observed at the time of vaccination in 22 patients. At the time of first vaccination, 58 patients were on immunosuppressants; 53 patients were on calcineurin inhibitors, 33 were on corticosteroids, and 7 were on mycophenolate mofetil. Specifically, calcineurin inhibitor alone was given to 24 patients and corticosteroid alone to five patients. Calcineurin inhibitor and corticosteroid were administered to 22 patients,calcineurin inhibitor and mycophenolate mofetil to 1 patient, and calcineurin inhibitor, corticosteroid, and mycophenolate mofetil to 6 patients. The median corticosteroid dose for patients undergoing treatment with prednisolone was 7 mg/day. The median lymphocyte count and IgG level at the first vaccination were 1.99×10^9/L and 956 mg/dl, respectively. Sixty-nine patients had a history of csCMVi, and the median interval from csCMVi to first vaccination dose was 1075 days (quantile, 357–1853). Nine patients underwent prophylactic treatment for primary hematological diseases at first vaccination with TKI for six patients and azacitidine for three patients.

The GMT of SARS-CoV-2 S1 Ab before vaccination was 0.62 AU/ml (median titer, 0.56 AU/ml). The titer was >10 AU/ml in two patients, specifically, 21.38 and 15.27 AU/ml. Because the SARS-CoV-2

TABLE 1 Clinical characteristics of allogeneic stem cell transplantation (SCT) recipients (n = 128)

Characteristic	n = 128
Patient age at vaccination, years	
Median age (quantile)	54 (46–63)
>50	75
≤50	53
Interval between SCT and vaccination	
>1 year	103
≤1 year	25
Patient sex	
Male	72
Female	56
Female to male	
Yes	22
No	106
History of prior allogeneic SCT	
No	118
Yes	10
Underlying disease	
Acute myeloid leukemia	51
Chronic myeloid leukemia	7
Myelodysplastic syndromes	27
MDS/MPN	2
Myelofibrosis	3
Acute lymphoblastic leukemia	22
T-lymphoblastic leukemia/lymphoma	2
Adult T-cell leukemia/lymphoma	1
Malignant lymphoma	4
Mixed phenotype acute leukemia	4
Aplastic anemia	5
Graft source	
Bone marrow	76
Peripheral blood	35
Cord blood	17
HLA	
Matched	64
Mismatched	64
Haploidentical SCT	14
Relationship	
Related	34
Unrelated	94
Blood type	
Matched	51
Mismatched	77
Conditioning intensity	
Myeloablative	72
Reduced intensity	56

TABLE 1 (Continued)

Characteristic	n = 128
TBI-containing conditioning regimen	
Yes	96
No	32
ATG use before SCT	
Yes	31
No	97
History of acute GVHD, grade	
0–1	81
2–4	47
Chronic GVHD at vaccination	
No	89
Mild	17
Moderate	14
Severe	8
Use of any immunosuppressant	
Yes	58
No	70
Use of calcineurin inhibitor at vaccination	
Yes	53
No	75
Use of corticosteroid at vaccination	
Yes	33
No	95
Use of MMF at vaccination	
Yes	7
No	121
Treatment for primary hematological disease at vaccination	
Yes	9
No	119
WBC count before vaccination	
>6000/μl	63
≤6000/μl	65
Neutrophil count before vaccination	
>3000/μl	76
≤3000/μl	52
Lymphocyte count before vaccination	
>2000/μl	64
≤2000/μl	64
IgG level before vaccination	
>900 mg/dl	68
≤900 mg/dl	49
NA	11
History of csCMVi	
Yes	69
No	59

Abbreviations: ATG, antithymocyte globulin; csCMVi, clinically significant CMV infection; GVHD, graft-versus-host disease; HLA, human leukocyte antigen; MDS/MPN, myelodysplastic/myeloproliferative neoplasm; MMF, mycophenolate mofetil; NA, not assessed; TBI, total body irradiation; WBC, white blood cell.

Ab test result for these two patients was negative, we included these patients in subsequent analyses for SARS-CoV-2 S1 Ab titers after vaccination. The GMT of SARS-CoV-2 S1 Ab 2 weeks after the first vaccination dose, that is, before the second vaccination dose, was 6.82 AU/ml (median, 4.28 AU/ml).

3.2 | Antibody titers after second vaccination

The SARS-CoV-2 S1 Ab became positive 2 weeks after the second vaccination dose in 87.6% of the SCT recipients, and the GMT was 456.48 AU/ml (median, 1235.4 AU/ml) (Figure 1A). The association between the Ab titers after the second vaccination dose and the clinical parameters was analyzed. The association of Ab titers with patients' age, lymphocyte count, and IgG levels at the first vaccination showed a correlation tendency (Figure 1B–D). Grouped comparison by clinical parameters revealed that a short interval from SCT, steroid use, and ATG treatment before SCT were critical factors for lower Ab titers (Figure 1E–G). Stem cell transplantation time, and moderate to severe cGVHD at vaccination were also associated with Ab titers (Figure S1A–G). In particular, the corticosteroid dose at the first vaccination inversely correlated with the Ab titer in all patients (rho = −0.48, $p < 0.0001$; Figure S1H), and the correlation was significant in 33 patients who were treated with corticosteroids (rho = −0.36, $p = 0.038$; Figure 1H). We noted that the Ab titer in nine patients who were treated with TKI or azacitidine at the time of the first vaccination seemed comparable with that of other patients, although the number was limited (Figure S1G). Furthermore, SARS-CoV-2 S1 Ab titer according to the combination of immunosuppressive agents was also evaluated, although the number of patients in some subgroups was low (Figure S2). The titer 2 weeks after the second vaccination dose was significantly higher in patients treated with calcineurin inhibitor alone than in those treated either with a combination of calcineurin inhibitor and corticosteroid or a combination of calcineurin inhibitor, corticosteroid, and mycophenolate mofetil (median 964.22 vs. 213.12 vs. 265.94 AU/ml, $p = 0.049$ and $p = 0.0045$).

Multivariate analysis identified that the interval from SCT (<1 year, OR 0.0840; 95% CI, 0.00709–1.00; $p = 0.050$), history of csCMVi (OR 0.114; 95% CI, 0.0143–0.908; $p = 0.040$), and steroid use (prednisolone >5 mg/day, OR 0.0567; 95% CI, 0.00502–0.640; $p = 0.020$) were risk factors for failure of positive conversion of SARS-CoV-2 S1 IgG Ab (Table 2).

3.3 | Diminishing vs. persistent Ab titers 6 months after vaccination

The GMT of SARS-CoV-2 S1 Ab 3 months after the first vaccination dose was 253.74 AU/ml (median, 581.70 AU/ml). The SARS-CoV-2 S1 Ab was positive in 83.6% of SCT recipients 6 months after first vaccination. However, the Ab titer significantly and gradually decreased after the second vaccination dose (Figure 2A), with the GMT reaching

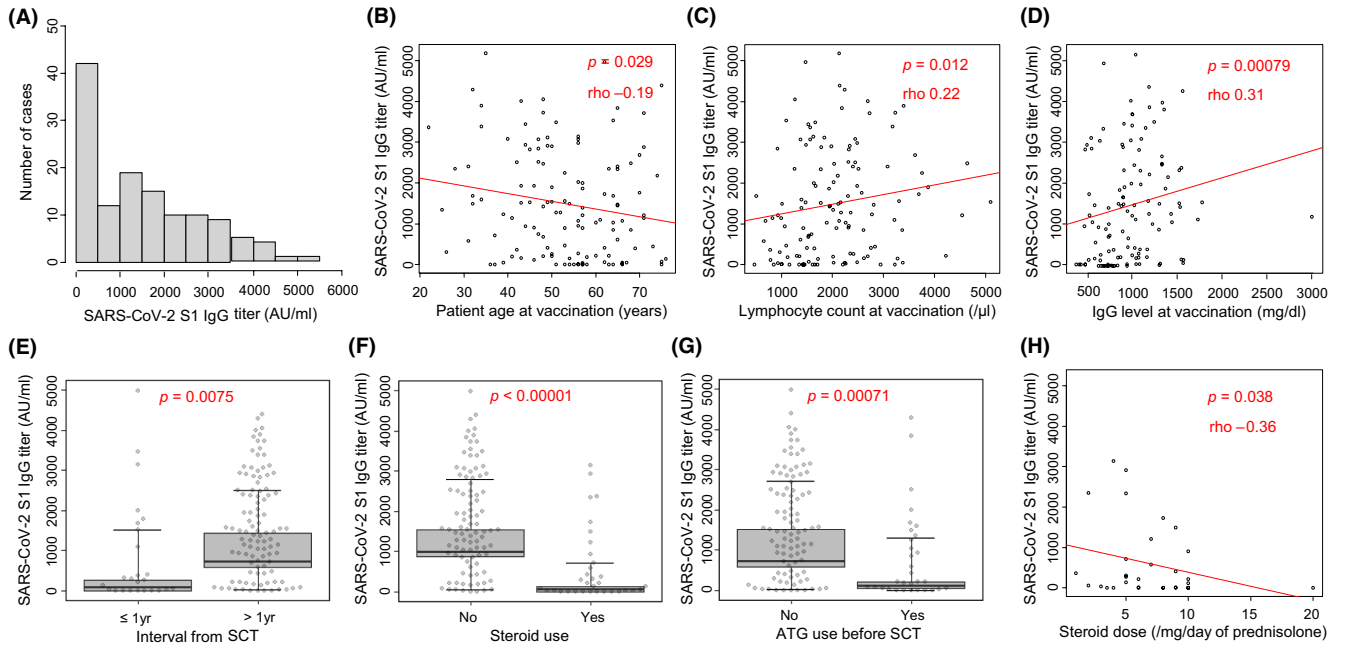

FIGURE 1 Clinical characteristics and severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) S1 Ab titers after second vaccination in allogeneic stem cell transplantation (SCT) recipients. (A) Histogram of SARS-CoV-2 S1 Ab titers. (B–D) Plot of SARS-CoV-2 S1 Ab titers and (B) patient age at vaccination, (C) peripheral blood lymphocyte counts at first vaccination, and (D) IgG levels at first vaccination. (E–G) Boxplot of SARS-CoV-2 S1 Ab titers and (E) interval from SCT to first vaccination, (F) use of corticosteroid at the time of first vaccination, and (G) administration of antithymocyte globulin (ATG) before SCT. (H) Plot of SARS-CoV-2 S1 Ab titers after second vaccination and dose of corticosteroid at first vaccination (mg/day of prednisolone).

82.54 AU/ml (median, 185.15 AU/ml), less than one-fifth of the level directly after the second vaccination dose. Antibody titers 6 months after vaccination were generally well correlated with those after the second vaccination dose (rho = 0.813, $p < 0.0001$; Figure 2B). In a separate analysis of the elapsed time from the SCT, the titers after the second vaccination dose of the over-1-year group were significantly higher than those of the 1-year-or-less group. However, this association was lost as time passed (Figure S3). Determinants for SARS-CoV-2 S1 IgG Ab titers after the second vaccination dose and at 6 months and their correlations, established through univariate analyses, are shown in Figure 2C to clarify differences in short- and long-term effects. Similar to the interval from the SCT, for some clinical factors the association with Ab titers at 6 months was no longer significant (Figure 2C).

Multivariate analysis revealed that steroid use (prednisolone >5 mg/day) was significantly negatively associated with the SARS-CoV-2 S1 IgG Ab titers 6 months after first vaccination (OR 0.0535; 95% CI, 0.00558–0.513; $p = 0.011$) (Table 3). The median SARS-CoV-2 S1 IgG Ab titers at 6 months were 38.6 in patients treated with prednisolone >5 mg/day versus 413.4 AU/ml in those not undergoing prednisolone treatment ($p < 0.0001$).

3.4 | Higher Ab titers after second vaccination

Considering the natural waning of Abs, a higher Ab titer 2 weeks after the second vaccination dose could be beneficial for persistent humoral immunity. The optimal cut-off was unknown; thus, we set SARS-CoV-2 S1 IgG > 500 AU/ml as the cut-off value because all patients with levels >500 AU/ml after the second vaccination dose remained SARS-CoV-2 S1 IgG positive 6 months after first vaccination. The SARS-CoV-2 S1 Ab titer 2 weeks after the second vaccination dose was >500 AU/ml in 86 patients (67.2%). Clinical factors associated with SARS-CoV-2 S1 IgG > 500 AU/ml 2 weeks after the second vaccination were quite different from those associated with levels >10 AU/ml (Figure 2C,D). The multivariate analysis showed that the interval from SCT (<1 year, OR 0.105; 95% CI, 0.0275–0.401; $p = 0.00097$), ATG-based haploidentical SCT (OR 0.0956; 95% CI, 0.0132–0.692; $p = 0.020$), and steroid use at vaccination (prednisolone >5 mg/day, OR 0.123; 95% CI, 0.0267–0.570; $p = 0.0074$) were risk factors for lower Ab titers (Table S1).

3.5 | Adverse effects after vaccination

Questionnaires were administered to 74 and 75 patients after the first and second vaccination doses, respectively. At least one adverse effect was observed in 85.1% and 80.0% of patients after the first and second vaccination doses, respectively. Local pain was the most common side-effect (Figure 3, Tables S2 and S3). Three (4.1%) and five (6.7%) patients reported deterioration of GVHD symptoms after the first and second vaccination doses, respectively, although none required starting or increasing systemic immunosuppressive therapy. No severe adverse effects were observed.

TABLE 2 Univariate and multivariate analyses for SARS-CoV-2 S1 Ab positivity after second vaccination

	Univariate analysis		Multivariate analysis	
Characteristics	**Odds ratio (95% CI)**	***p* value**	**Odds ratio (95% CI)**	***p* value**
IgG > 900 mg/dl	13.9 (2.97–132)	<0.0001	6.31 (0.489–81.5)	0.16
Interval from SCT (≤1 year)	5.95 (1.78–20.4)	0.0014	0.0840 (0.00709–1.00)	0.05
Lym > 2000/μl	3.77 (1.08–16.8)	0.0350	2.89 (0.488–17.1)	0.24
Moderate to severe cGVHD	0.344 (0.100–1.28)	0.0840	0.237 (0.0179–3.15)	0.28
ATG use before SCT	0.316 (0.0970–1.05)	0.0350	0.262 (0.0387–1.78)	0.17
Patient age > 50 years	0.262 (0.0460–1.01)	0.0360	0.312 (0.0392–2.49)	0.27
History of csCMV infection	0.213 (0.0370–0.820)	0.0170	0.114 (0.0143–0.908)	0.04
Calcineurin inhibitor use	0.179 (0.0400–0.629)	0.0029	2.23 (0.242–20.7)	0.48
Rituximab use before SCT	0.172 (0.0260–1.29)	0.0450	0.264 (0.00318–22.0)	0.55
Prednisolone >5 mg/day	0.0600 (0.0150–0.216)	<0.0001	0.0567 (0.00502–0.640)	0.02

Note: IgG, lymphocyte count (Lym), status of chronic graft-versus-host disease (cGVHD), age, use of immunosuppressants were at the time of first vaccination.

Abbreviations: ATG, antithymocyte globulin; CI, confidence interval; csCMV, clinically significant CMV; SCT, stem cell transplantation.

3.6 | COVID-19 breakthrough infection after vaccination

Five patients were diagnosed with COVID-19 at least 2 weeks after the second vaccination, and the 300-day cumulative incidence of COVID-19 breakthrough infection was 7.4% (Figure S4). The median interval between the first vaccination dose and COVID-19 diagnosis was 231 days (range, 195–275 days). None of the patients received a third vaccination before COVID-19 diagnosis. The SARS-CoV-2 S1 Ab was positive in four of the five patients after the second vaccination dose, and the Ab titers were 4971.13, 2506.41, 1531.28, 64.5, and 0.43 AU/ml, respectively. The severity of COVID-19 was mild in three patients, including one without sufficient humoral response, and moderate in two. All patients improved after treatment. Management with sotrovimab was required for three patients, remdesivir for one, and regular observation for one. The variant of SARS-CoV-2 in each patient was not evaluated. The timing of breakthrough infection was January 2022 for one patient, February 2022 for three patients, and April 2022 for one patient.

4 | DISCUSSION

In this study, humoral response was achieved in more than 85% of SCT recipients after two doses of the SARS-CoV-2 mRNA vaccine. This is the largest study to evaluate humoral response of mRNA vaccine in Japanese allogeneic SCT recipients, and the response rate was comparable with that reported in previous studies.[9–11,13,18–20] These studies have identified that some clinical characteristics, such as lymphocytopenia, hypoglobulinemia, and a shorter interval from the SCT, were associated with a poor response. We further clarified that treatment with corticosteroid was important for poor humoral response and that the dose of corticosteroid also had a significant impact.

We also showed that many of the parameters were not significantly associated with the persistence of the humoral response 6 months after first vaccination. Leclerc et al. also revealed that a lower peak titer, use of immunosuppressive medications, and lymphocytopenia predict a poorly persisting response,[12] which is consistent with our results. In Japan, a third vaccination is recommended at least 6 months after the second dose regardless of the immune status, although previous studies in Europe and the United States underwent earlier additional vaccination for SCT recipients.[21–23] Early booster shots should be effective for potent humoral response, and optimization of additional vaccination schedules for immunocompromised hosts based on appropriate evaluation of humoral response could contribute to better infection control and a possible preventive role for the advent of novel variants.[24]

Interestingly, a history of csCMVi was associated with a poor humoral response in this study. These results might reflect impaired immunity in the recipients. However, CMV reactivation is associated with modified immunity[25–28] and variable infections.[29–31] In addition, CMV reactivation has been reported to be associated with a poor response after vaccination against hepatitis B virus in SCT recipients.[32] These reports suggest an association between CMV infection and impaired acquired immunity against other viruses approximately 1 year later. The median interval from csCMVi to vaccination in our cohort was approximately 3 years; therefore, the possible immunological impact of CMV reactivation lasts for a few years, although further validation using other cohorts and elucidation of the underlying mechanisms are warranted.

The adverse effects of vaccination in SCT recipients were generally mild in this cohort. Adverse effects in SCT recipients have been reported to be generally tolerable.[33–35] Nonetheless, careful follow-up is warranted because severe adverse effects after SARS-CoV-2 vaccination could occur in SCT recipients.[36,37]

Five breakthrough infections were noted in our study, and all occurred more than 6 months after the first vaccination dose.

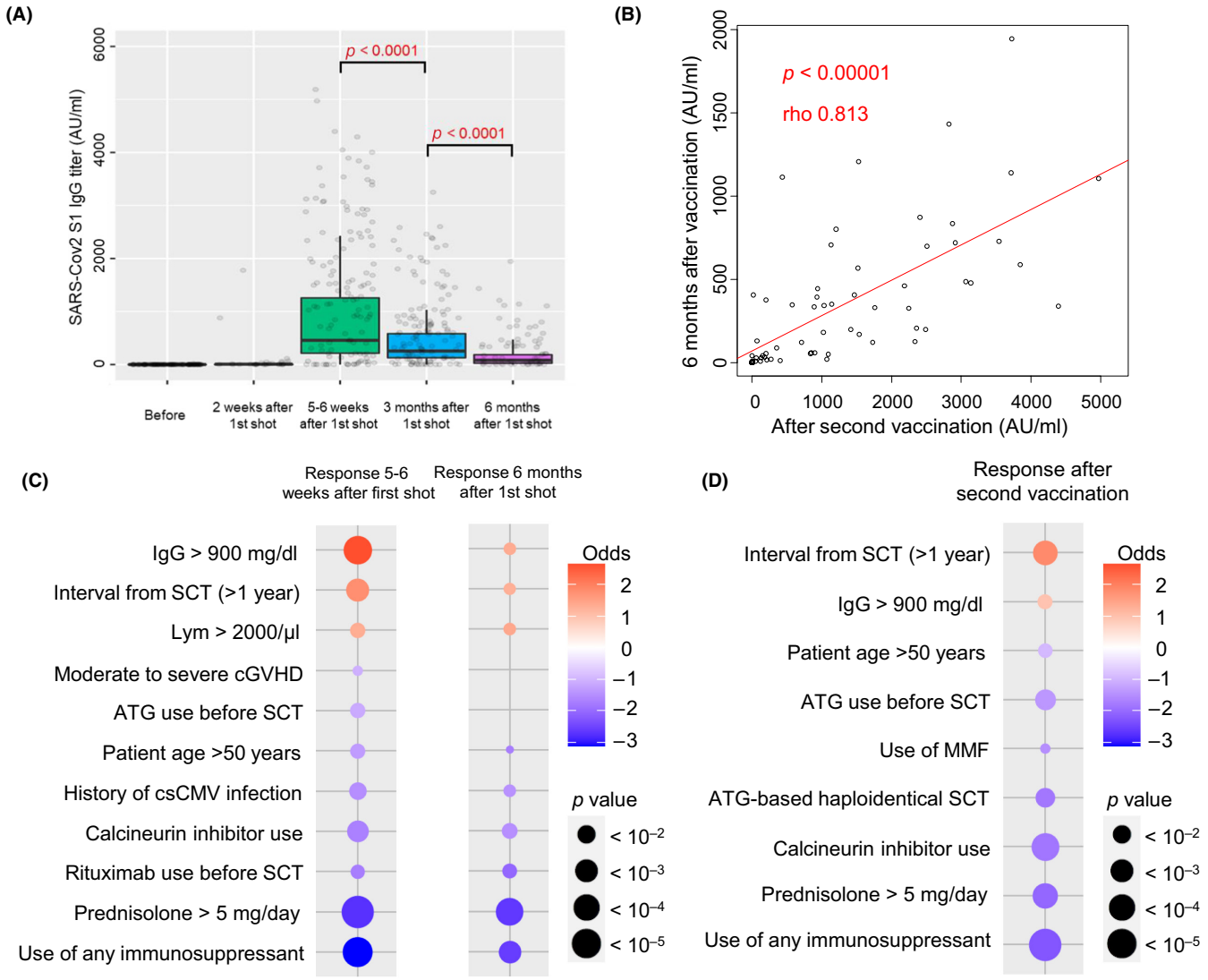

FIGURE 2 Clinical characteristics associated with severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) S1 Ab titers and their persistence in allogeneic stem cell transplantation (SCT) recipients. (A) Box plot of transition of SARS-CoV-2 S1 Ab titer. (B) Plot of SARS-CoV-2 S1 Ab titers after second vaccination and 6 months after vaccination. (C) Univariate analyses of SARS-CoV-2 S1 Ab >10 AU/ml after second vaccination (left column) and 6 months after vaccination (right column) and clinical parameters. Characteristics with p value <0.1 are shown. The color of circles indicates odds ratio and the size indicates p value. (D) Univariate analyses of SARS-CoV-2 S1 Ab >500 AU/ml after second vaccination. ATG, antithymocyte globulin; AU, arbitrary unit; csCMV, clinically significant CMV; cGVHD, chronic graft-versus-host disease; Lym, lymphocyte count; MMF, mycophenolate mofetil.

Unfortunately, variants of SARS-CoV-2 were not identified, in part because the variant in each patient was not routinely evaluated in Japan during the study period. However, considering the timing of infection and the epidemic strains in Tokyo at the time,[38] all patients were considered to be infected with the Omicron strain and, specifically, BA1.1 in four cases and BA.2 in one case. The exact contribution of the characteristics of the infected strains and the duration from second vaccination was uncertain. However, a third vaccination dose as soon as 6 months after the second vaccination dose seems a reasonable option considering that the Ab titer 6 months after vaccination appeared to be insufficient to prevent Omicron variant breakthrough infection[39] and that humoral immunity against variants of concern including the Omicron strain can be enhanced with a third dose.[40,41]

Our study had several limitations. First, cellular immunity was not analyzed in this study. Some studies analyzed cellular immunity against COVID-19 in immunocompromised hosts,[23,42–45] and, as Harrington et al. reported, cGVHD and ongoing immunosuppressive therapy were associated with a reduced T cell response.[44] Further analyses identifying factors associated with the response and persistence of cellular immunity in SCT recipients are warranted. Second, neutralizing Abs were not analyzed. However, previous reports showed that the SARS-CoV-2 S1 Ab titer correlated well with neutralizing Ab titers[46]; therefore, we considered that the SARS-CoV-2 S1 Ab titer was a useful and practical marker in assessing SARS-CoV-2 humoral immunity. Third, our analyses of adverse effects were based on a questionnaire survey and thus

TABLE 3 Univariate and multivariate analysis for SARS-CoV-2 S1 Ab positivity after 6 months

	Univariate analysis		Multivariate analysis	
Characteristic	**Odds ratio (95% CI)**	***p* value**	**Odds ratio (95% CI)**	***p* value**
IgG > 900 mg/dl	3.81 (0.850–24.0)	0.0640	1.13 (0.111–11.5)	0.920
Interval from SCT (≤1 year)	3.58 (0.821–15.5)	0.0670	0.463 (0.417–5.15)	0.530
Lym > 2000/μl	4.02 (0.910–25.1)	0.0630	2.64 (0.305–22.8)	0.380
History of csCMV infection	0.228 (0.0230–1.19)	0.0630	0.146 (0.0115–1.85)	0.140
Calcineurin inhibitor use	0.216 (0.0350–0.958)	0.0310	0.800 (0.0883–7.24)	0.840
Rituximab use before SCT	0.125 (0.00900–1.23)	0.0400	0.0329 (0.000325–3.34)	0.150
Prednisolone >5 mg/day	0.0650 (0.0110–0.304)	<0.0001	0.0535 (0.00558–0.513)	0.011

Note: IgG, lymphocyte count (Lym), use of immunosuppressants were at the time of first vaccination.
Abbreviations: CI, confidence interval; csCMV, clinically significant CMV; SCT, stem cell transplantation.

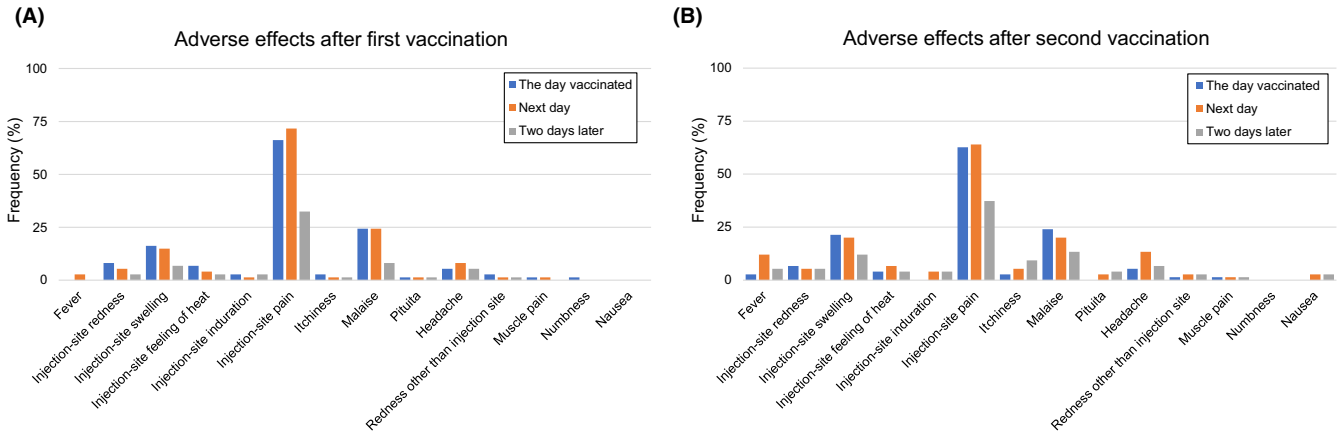

FIGURE 3 Adverse effects in allogeneic stem cell transplantation recipients after (A) first and (B) second severe acute respiratory syndrome coronavirus 2 vaccination.

objective validity was lacking. In addition, the response rate was low, which resulted in a possible selection bias. However, previous studies involving SCT recipients reported a comparable incidence of adverse effects after SARS-CoV-2 mRNA vaccination[35,47]; therefore, we considered that our results had acceptable validity. Finally, there was some variation in the vaccination schedule and timing of sample collection, and some serial samples were unavailable for some patients. The unreliable and unpredictable supply of vaccines combined with the vulnerable medical system prevented us from undertaking punctual sampling, which might have affected the results.

In conclusion, a positive humoral response after two vaccination doses was observed in 87.6% of SCT recipients. A higher dose of corticosteroid given around the time of vaccination was associated with a poor response. A shorter interval after SCT, history of csCMVi, and steroid use were significantly associated with SARS-CoV-2 S1 IgG < 500 AU/ml after the second vaccination dose. Steroid use was also associated with a lack of persistent response. Finally, the SARS-CoV-2 mRNA vaccine appears to be safe for patients with SCT.

AUTHOR CONTRIBUTIONS

Takashi Toya, Daichi Sadato, Noritakaa Sekiya, and Michinori Kohara designed the study. Takashi Toya, Yuya Atsuta, Naoki Shingai, Daishi Onai, Naoki Shingai, Hiroaki Shimizu, Yuho Najima, Takeshi Kobayashi, Kazuteru Ohashi, and Noriko Doki provided medical treatment, obtained informed consent, and collected the clinical data. Daichi Sadato, Hiroko Kogure, Sonomi Takakuwa, and Yuka Harada collected and managed clinical samples. Takahiro Sanada, Tomoko Honda, and Michinori Kohara analyzed the Ab titers. Takashi Toya and Daichi Sadato performed the statistical analyses, wrote the manuscript, and created the figures. Kazuteru Ohashi, Yuka Harada, and Noriko Doki supervised this study. All authors have read and approved the manuscript.

ACKNOWLEDGMENTS

We thank all the patients who participated in this study. We thank Fumihiko Yasui for his kind and valuable suggestions regarding the study design, and Junko Ohta and Atsuko Yoshida for attentive sample management. We also thank the medical and laboratory staff at Tokyo Metropolitan Komagome Hospital. We would like to thank Editage for English language editing.

DISCLOSURE

The authors declare no conflict of interest.

ETHICS STATEMENT

Approval of the research protocol by an institutional review board: This study was performed in accordance with the Declaration of Helsinki and approved by the Ethics Committee of Tokyo Metropolitan Komagome Hospital (approval number 2716).

Informed consent: Written informed consent was obtained from all the patients.

Registry and registration no. of the study/trial: N/A.

Animal studies: N/A.

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SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

How to cite this article: Toya T, Atsuta Y, Sanada T, et al. Attenuated humoral response against SARS-CoV-2 mRNA vaccination in allogeneic stem cell transplantation recipients. *Cancer Sci*. 2023;114:586-595. doi: 10.1111/cas.15603