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Humoral and T-cell–mediated immunity to SARS-CoV-2 vaccination in patients with liver disease and transplant recipients

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Abstract

Background: SARS-CoV-2 vaccination induces a varied immune response among persons with chronic liver disease (CLD) and solid organ transplant recipients (SOTRs). We aimed to evaluate the humoral and T-cell–mediated immune responses to SARS-CoV-2 vaccination in these groups.

Methods: Blood samples were collected following the completion of a standard SARS-CoV-2 vaccination (2 doses of either BNT162b2 or mRNA-12732), and a subset of patients had a blood sample collected after a single mRNA booster vaccine. Three separate methods were utilized to determine immune responses, including an anti-spike protein antibody titer, neutralizing antibody capacity, and T-cell–mediated immunity.

Results: The cohort included 24 patients with chronic liver disease, 27 SOTRs, and 9 controls. Patients with chronic liver disease had similar immune responses to the wild-type SARS-CoV-2 compared with controls following a standard vaccine regimen and single booster vaccine. SOTRs had significantly lower anti-S1 protein antibodies ($p < 0.001$), neutralizing capacity ($p < 0.001$), and T-cell–mediated immunity response ($p = 0.021$) to the wild-type SARS-CoV-2 compared with controls following a standard vaccine regimen. Following a single booster vaccine, immune responses across groups were not significantly different but numerically lower in SOTRs. The neutralization capacity of the B.1.1.529 Omicron variant was not significantly different between groups after a standard vaccine regimen ($p = 0.87$) and was significantly lower in the SOTR group when compared

Abbreviations: BAU, binding antibody unit; CLD, chronic liver disease; CMI, cell-mediated immunity; HC, healthy controls; IFN-γ, interferon-gamma; IQR, interquartile range; IS, immunosuppressed; MMF, mycophenolate mofetil; N, number; RBD, receptor binding domain; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; SFU, spot forming unit; SOTR, solid organ transplant recipient; WHO, World Health Organization; WT, wild type

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with controls after a single booster vaccine ($p = 0.048$).

Conclusion: The immunogenicity of the SARS-CoV-2 vaccine is complex and multifactorial. Ongoing and longitudinal evaluation of SARS-CoV-2 humoral and cellular responses is valuable and necessary to allow frequent re-evaluation of these patient populations.

INTRODUCTION

Vaccination against the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) virus is a mainstay of prevention and mitigation of COVID-19, especially in vulnerable and at-risk populations. Recent guidelines from liver societies recommend vaccination against SARS-CoV-2 in persons with chronic liver disease, liver transplant candidates, and liver transplant recipients.[1,2] Although patients with chronic liver disease (CLD) were eligible for enrollment in the initial SARS-CoV-2 vaccine licensing trials, the sample size was limited and subgroup analyses were not performed.[1] There are very limited data on the humoral and cellular responses of SARS-CoV-2 vaccination in patients with CLD in the literature. In posttransplant, immunosuppressed cohorts, there are several reports aimed at evaluating the vaccine immune responses and the effects of multidose regimens, but few studies include data regarding immune response to the Omicron variants.[3–5]

Published data suggest that immune response to SARS-CoV-2 vaccination among persons with CLD varies and is less robust in solid organ transplant recipients (SOTRs) that are immunosuppressed (IS), especially in older persons and those receiving higher doses of immunosuppressive therapy, compared with that of the general population.[6–12] Administration of a booster vaccine yields a favorable humoral response in these populations.[8,9,13,14] However, despite the detectable antibody response, concern remains about the neutralization capacity of vaccine-induced immunity as new COVID-19 variants arise and spread. Lastly, there is limited literature that discusses the T-cell–mediated immune responses in these populations following SARS-CoV-2 vaccination.

We aimed to evaluate the humoral and T-cell–mediated immune responses to vaccination against SARS-CoV-2 in patients with CLD and SOTRs compared with the healthy controls following a standard vaccine regimen. We also evaluated a subset of patients for humoral immune response following a single booster vaccine. We utilized 3 separate methods to quantify the immune responses, including the anti-spike (S1) antibody titer, the neutralizing antibody capacity of the wild-type SARS-CoV-2 and the B.1.1.529 Omicron variant, and the T-cell–mediated responses to spike epitopes as determined by interferon-gamma (IFN-γ) ELISPOT assay.

METHODS

Study design and patient population

Adults aged 18 years and older were enrolled in a biobank repository study approved by the Institutional Review Board at the University of Cincinnati, and all subjects provided informed consent. This cohort included healthy, immunocompetent controls, patients with chronic liver disease with and without cirrhosis, and solid organ transplant recipients (liver, or liver and kidney).

We collected clinical data from the patients' medical records, including demographic data and confirmation of the vaccination type and date. The etiology of liver disease was obtained for CLD. Patients with cirrhosis were identified by either a liver biopsy or stage 4 liver fibrosis on transient elastography. The date of transplant and the use of immunosuppressive medications were collected for SOTRs.

Serum/plasma and whole blood samples containing peripheral blood mononuclear cells were collected following vaccination with the standard regimen of an Emergency Use Authorization-approved SARS-CoV-2 vaccine. A standard regimen for SARS-CoV-2 vaccination was defined as 2 doses of an mRNA vaccine (either BNT162b2 manufactured by Pfizer/BioNTech or mRNA-1273 manufactured by Moderna). Blood samples were also collected after receiving a single mRNA booster vaccine against SARS-CoV-2 from either Pfizer or Moderna following the completion of their standard vaccine regimen.

Exclusion criteria included patients with a documented positive test for SARS-CoV-2 infection, a positive nucleocapsid test, or an incomplete standard vaccine regimen. Patients who received the Johnson & Johnson/Janssen vaccine (Ad26.COV2.S) and/or booster vaccine were also excluded.

Serum/plasma and whole blood samples were identified from the biobank repository based on the study criteria (Supplemental Fig. 1, http://links.lww.com/HC9/A208). Then, the humoral and T-cell–mediated immune responses were retrospectively collected from the available patient samples. The known number (N) was reported to identify incomplete data within this cohort.

Evaluation of immune response

For humoral immune response, anti-spike (S1) protein antibody titers were evaluated using a quantitative ELISA assay (anti-SARS-CoV-2 QuantiVac ELISA (IgG), EUROIMMUN US Inc., NJ, USA) that determined the concentration of IgG antibodies against the S1 domain of the spike protein of SARS-CoV-2 reported in WHO binding antibody units (BAU). A positive cutoff of 35.2 BAU was utilized as recommended by the manufacturer. All samples were diluted at 1:100, and any above the linear range was further diluted.

Neutralizing antibody capacity was quantified by the ability of the antibody to block binding to the SARS-CoV-2 receptor binding domain (RBD) of the ACE-2 receptor (SARS-CoV-2 Surrogate Virus Neutralization Test (sVNT) Kit (RUO), also known as cPass^{TM} SARS-CoV-2 Neutralization Antibody Detection Kit, GenScript USA Inc., NJ, USA). This test detects and measures circulating neutralizing antibodies against the wild-type SARS-CoV-2 virus. The percent signal inhibition was determined by the equation

$$\left(1 - \frac{\text{sample OD value}}{\text{negative control OD value}}\right) \times 100\%.$$

A positive cutoff of 30% was utilized as recommended by the manufacturer. By replacing the protein used in the assay with SARS-CoV-2 spike protein RBD-HRP B.1.1.529 Omicron variant, neutralizing antibodies to the Omicron RBD were determined.

To evaluate the T-cell–mediated immunity (CMI), IFN-γ ELISPOT to SARS-CoV-2 S1 peptides was performed as described.[15] Using peripheral blood mononuclear cells separated from whole blood, peptide pools covering the N-terminal S1 domain of the spike glycoprotein of the wild-type SARS-CoV-2 were used for cell stimulation (PepTivator SARS-CoV-2 Prot_S1, Miltenyi Biotec, Cambridge, MA). The number of SARS-CoV-2–specific IFN-γ–secreting cells was estimated using the CTL ImmunoSpot S6 Analyzer (Cleveland, OH, USA) to detect spots with predetermined criteria based on the size, shape, and colorimetric density. Quantification of the SARS-CoV-2–specific T-cell response was calculated by subtracting the number of IFN-γ–secreting cells given no stimulation (medium alone) from the number of IFN-γ–secreting cells induced by stimulation of the test well for a given sample. This was expressed as spot forming units per 10^6 cells. The cutoff for a positive ELISPOT response was determined using participant samples obtained before vaccination or a known COVID infection determined by the history or presence of a nucleocapsid antibody. The cutoff was calculated using the upper limit of the 95% CI of the stimulated minus unstimulated response of this true negative group and was determined to be 46 spot forming units/10^6 cells.

Statistical analysis

Categorical variables were reported as the number of patients and percentage. Continuous variables were reported as the mean and SD for normally distributed data and median and interquartile range (IQR) for non-normally distributed data. Categorical variables were compared with either chi-square or Fisher exact test as appropriate. Non-normally distributed continuous variables were compared using a Mann-Whitney *U* test for 2 groups or a Kruskal-Wallis test and associated pairwise comparison with Bonferroni correction for more than 2 groups. A Wilcoxon signed-rank test was used for the comparison of paired and non-normally distributed data when $N \geq 5$. Statistix version 10 (Analytical Software, Tallahassee, FL) and SPSS Statistics version 28 (IBM Corp, Armonk, NY) were used for statistical analyses. A value of $p < 0.05$ was considered statistically significant for all analyses based on known data.

RESULTS

Patient characteristics

The demographic and clinical characteristics of the study cohort are described in Table 1. Blood/serum samples were collected from 60 patients, including 9 healthy controls, 24 patients with CLD, and 27 solid organ transplant recipients. Seven (29%) of the CLD patients had cirrhosis. There were 27 solid organ transplant recipients, including 26 liver transplant recipients and 1 combined liver and kidney transplant recipient. The majority of the liver transplant recipients in our cohort were on an immunosuppressive regimen with tacrolimus and mycophenolate mofetil (N = 17, 63%) or tacrolimus monotherapy (N = 5, 19%). Overall, the mean (SD) age was 58.6 ± 11.3 years. About half of the study cohort were male (57%, N = 34), and the majority were non-Hispanic white (82%, N = 49).

Fifty-two blood/serum samples were analyzed from participants who received the standard vaccine regimen (2 doses of either Pfizer or Moderna), and 19 samples were from patients who received a single booster vaccine. The median number of days after the completion of a standard vaccine regimen at the time of blood sample collection was 31 (range 23–103) days. Nineteen of the 60 patients received a single booster vaccine, and these subsequent samples were collected at a median of 62 (range 40–91) days after the booster.

Anti-S1 protein antibody response to wild-type SARS-CoV-2

Anti-S1 antibody responses to the wild-type SARS-CoV-2 are summarized in Table 2. Following a standard

TABLE 1 Demographic and clinical characteristics of the study cohort stratified patient group

Patient characteristics	Overall (N = 60)	Controls (N = 9)	CLD (N = 24)	SOTR (N = 27)
Age, y (mean, SD)	58.6 ± 11.3	51.0 ± 14.5	61.0 ± 9.0	58.9 ± 11.4
Male sex (N, %)	34 (57)	2 (22)	13 (54)	19 (70)
Vaccine manufacturer for standard regimen (N, %)				
Pfizer/BioNTech (BNT162b2)	31 (52)	0 (0)	13 (54)	18 (67)
Moderna (mRNA-1273)	29 (48)	9 (100)	11 (46)	9 (33)
Days after the completion of standard vaccine regimen (median, IQR)	31 (23–103)	28 (22–164)	36 (16–104)	30 (24–79)
Vaccine manufacturer for booster vaccine (N, %)	N = 19	N = 7	N = 4	N = 8
Pfizer/BioNTech	8 (42)	0 (0)	3 (75)	5 (63)
Moderna	11 (58)	7 (100)	1 (25)	3 (27)
Days after the completion of booster vaccine (median, IQR)	62 (40–91)	61 (53–78)	46 (16–173)	77 (41–96)
Etiology of liver disease (N, %)	—	—	—	—
Viral	—	—	15 (63)	—
Alcohol-associated liver disease	—	—	3 (13)	—
Autoimmune liver disease	—	—	4 (16)	—
NAFLD/NASH	—	—	1 (4)	—
Hereditary hemorrhagic telangiectasia	—	—	1 (4)	—
Cirrhosis present (N, %)	—	—	7 (29)	—
Maintenance immunosuppression for CLD patients (N, %)				
Azathioprine only	—	—	1 (4)	—
Azathioprine + steroid	—	—	2 (8)	—
Maintenance immunosuppression for SOTR				
Tacrolimus only	—	—	—	5 (19)
Tacrolimus + MMF	—	—	—	17 (63)
Tacrolimus + MMF + steroid	—	—	—	3 (11)
Other[a]	—	—	—	2 (7)
Days after transplant at time of standard vaccine (median, IQR)	—	—	—	550 (218–1050)
Days after transplant at time of booster vaccine (median, IQR)	—	—	—	1189 (400–2086)

[a]Other maintenance immunosuppressive regimens include tacrolimus/everolimus/prednisone and everolimus/MMF.
Abbreviations: CLD indicates chronic liver disease; IQR, interquartile range; N, number; SOTR, solid organ transplant recipients; MMF, mycophenolate mofetil.

vaccine regimen, antibodies to SARS-CoV-2 spike antigen were significantly lower in SOTRs when compared with immunocompetent controls ($p < 0.001$) and patients with CLD ($p = 0.001$), as shown in Figure 1A. Antibodies to SARS-CoV-2 spike antigen were similar in patients with CLD when compared with immunocompetent controls ($P = 0.40$) though a trend toward lower titers was noted. All controls (N = 9) and 95% of CLD patients (N = 20 of 21) elicited a positive anti-S1 antibody response to a standard vaccine regimen compared to 41% of SOTRs (N = 9 of 22; Figure 1C).

Following a single SARS-CoV-2 booster vaccine in addition to the standard vaccine regimen, the spike antigen response approached but did not reach statistical significance between groups ($p = 0.067$). In SOTRs, the spike antibody response after a single booster vaccine also trended toward lower levels of protection compared to controls ($p = 0.070$; Figure 1B). All controls (N = 7) and patients with CLD (N = 4) had a 100% positive anti-S1 antibody response along with 75% of SOTRs (N = 6 of 8) following a single booster vaccine (Figure 1D).

A subgroup analysis of paired data from the healthy controls following a standard vaccine regimen and after a single booster vaccine demonstrated no significant difference in the antibody to SARS-CoV-2 spike antigen in controls ($P = 0.87$, N = 7). The subgroup analysis of paired data could not be performed in patients with CLD (N = 1) and SOTR (N = 3) due to N being less than 5 in each group.

Neutralization capacity of the wild-type SARS-CoV-2

The neutralization capacity of the wild-type SARS-CoV-2 in each group is summarized in Table 2. SOTR had a significantly lower neutralizing capacity of the wild-type

TABLE 2 Humoral and T-cell–mediated immune responses after the completion of a standard vaccine regimen and a single booster vaccine stratified by patient group

Immune response	Healthy controls	Chronic liver disease	Solid organ transplant recipients	*p*
Anti-S1 IgG to wild-type SARS-CoV-2 (WHO BAU)				
Standard vaccine regimen	1550.4 (386.3–7185.8), N = 9	369.2 (171.6–1359.3), N = 21	3.92 (0.0–149.8), N = 22	**< 0.001**
Single booster vaccine	3340.9 (1615.6–5847.3), N = 7	1132.2 (624.9–3854.8), N = 4	407.1 (21.3–2872.0), N = 8	0.067
Neutralization capacity of wild-type SARS-CoV-2				
Standard vaccine regimen	96.4% (95.6–96.7%), N = 9	93.7% (76.1–95.6%), N = 21	28.0 % (15.4–64.3 %), N = 17	**< 0.001**
Single booster vaccine	96.2 % (95.4–96.3 %), N = 7	96.2 % (87.2–96.7 %), N = 4	71.8 % (11.3–96.7 %), N = 8	0.47
Neutralization capacity of the B.1.1.529 Omicron variant				
Standard vaccine regimen	27.1% (13.5–33.62%), N = 7	17.9 % (13.2–40.0 %), N = 19	19.2% (10.1–22.6%), N = 14	0.87
Single booster vaccine	87.3% (58.0–88.1%), N = 7	49.9% (10.4–78.4%), N = 4	29.2% (23.4–59.4%), N = 8	**0.046**
T-cell–mediated IFN-γ ELISPOT release assay (SFU per 10^6 cells)				
Standard vaccine regimen	147.5 (42.1–240.6), N = 8	80.4 (15.2–256.3), N = 10	18.8 (3.1–56.3), N = 15	**0.015**

Note: All data reported as median and interquartile range. Significant values of $p < 0.05$ are bolded.
Abbreviations: IFN-γ indicates interferon-gamma; N, number; SFU, spot forming units; WHO BAU, World Health Organization binding antibody units.

SARS-CoV-2 when compared with controls and patients with CLD ($p < 0.001$ and $p = 0.023$, respectively) following a standard vaccine regimen (Figure 2A). The neutralizing capacity of the wild-type SARS-CoV-2 spike antigen was numerically lower but not significantly different in patients with CLD when compared with immunocompetent controls ($p = 0.091$). All controls (N = 9) had a positive neutralizing capacity of the wild-type SARS-CoV-2 after a standard vaccine regimen compared to 95% of patients with CLD (N = 20 of 21) and 47% of SOTRs (N = 8 of 17; Figure 2C).

After a single booster vaccine, there were no significant differences in the wild-type neutralizing capacity between groups ($p = 0.47$; Figure 2B). All controls (N = 7) and patients with CLD (N = 4) had 100% positive wild-type neutralizing capacity along with 75% of SOTRs (N = 6 of 8) following a single booster vaccine (Figure 2D).

A subgroup analysis of paired data from healthy controls following a standard vaccine regimen and after a single booster vaccine demonstrated no significant difference in the neutralizing capacity of the wild-type SARS-CoV-2 in controls ($p = 0.24$, N = 7). The subgroup analysis of paired data could not be performed in CLD (N = 1) and SOTRs (N = 3).

Neutralization capacity of the B.1.1.529 omicron variant

The neutralization capacity of the B.1.1.529 Omicron variant in each group is summarized in Table 2. The neutralizing capacity of the Omicron variant following a standard vaccine regimen was not significantly different between groups ($p = 0.87$; Figure 3A). Only 29% of healthy controls (N = 2 of 7), 26% of patients with CLD (N = 5 of 19), and 14% of SOTRs (N = 2 of 14 had a positive neutralizing capacity of the Omicron variant after a standard vaccine regimen (Figure 3C).

The neutralization capacity of the B.1.1.529 Omicron variant was significantly lower in the SOTR group when compared with controls ($p = 0.048$, Figure 2C). All controls (N = 7) and 75% of patients with CLD (N = 3 of 4) had a positive neutralizing capacity of the Omicron variant after a booster vaccine compared with 38% of SOTR (N = 3 of 8; Figure 3C).

A subgroup analysis of paired data from healthy controls following a standard vaccine regimen and after a single booster vaccine demonstrated a significantly higher neutralizing capacity of the B.1.1.529 Omicron variant in controls ($p = 0.018$, N = 7). The subgroup analysis of paired data could not be performed in CLD (N = 1) and SOTRs (N = 3).

T-cell–mediated response to the wild-type SARS-CoV-2 following a standard vaccine regimen

The IFN-γ ELISPOT assay was available on a separate subset of patients following the completion of a standard vaccine regimen (N = 33; Table 2 and Figure 4A). When compared with controls, there was no significant

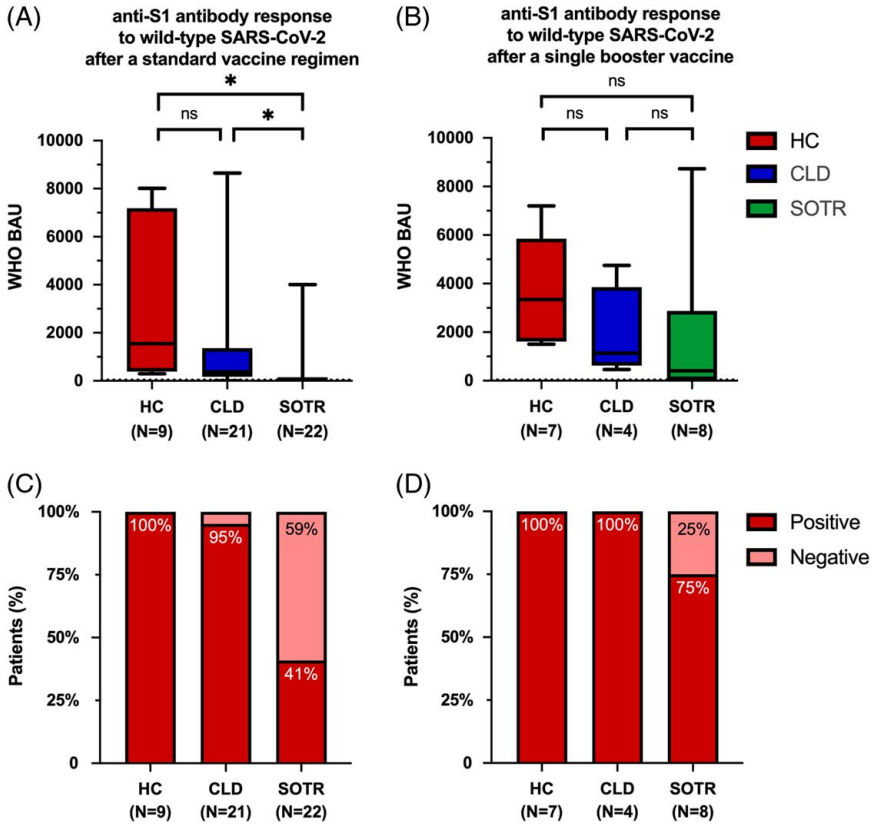

FIGURE 1 Evaluation of anti-S1 antibody response to wild-type SARS-CoV-2 after completion of a standard vaccine regimen (A and C) and following a booster vaccine (B and D) in each group. The boxplots represent the median and interquartile range with whiskers showing the minimum and maximum values. Dotted horizontal lines indicate the cutoff values for positive and negative responses. The respective proportions are provided as bar graphs. Abbreviations: CLD, chronic liver disease; HC, healthy controls; SOTR, solid organ transplant recipients; WHO BAU, World Health Organization binding antibody units. *$p < 0.05$.

difference in CMI in patients with CLD ($p = 0.99$), and the response was significantly lower in SOTRs ($p = 0.021$). Seventy-five percent of the healthy controls (N = 6 of 8) had a positive CMI along with 60% of patients with CLD (N = 6 of 10) and only 33% of SOTRs (N = 5 of 15; Figure 4B).

Immune response in patients with cirrhosis

A subgroup analysis of immune response in CLD patients with cirrhosis compared with the CLD patients without cirrhosis demonstrated that immune responses were similar after a standard vaccine regimen (Supplemental Table 1, http://links.lww.com/HC9/A209). A comparison of immune responses after a single booster vaccine was not performed due to the small sample size (N < 5).

Overall immune responses to the wild-type SARS-CoV-2 after a standard vaccine regimen

Seropositivity in all the 3 tests (anti-S1 protein antibody, antibody neutralization capacity, and T-cell–mediated ELISPOT) after a standard vaccine regimen was evaluated in 28 patients (8 HC, 10 CLD patients, 10 SOTRs) as shown in Figure 5. Most controls (N = 6, 75%) and patients with CLD (N = 6, 60%) demonstrated seropositivity in all the 3 tests. Only 20% (N = 2) of SOTRs were seropositive in all the 3 tests, and 20% (N = 2) did not elicit an immune response in any of the 3 tests.

DISCUSSION

We evaluated the humoral and T-cell–mediated immune responses to the SARS-CoV-2 vaccine, quantified by 3 separate methods, in patients with CLD and immunosuppressed patients/solid organ transplant recipients. Overall, immune responses to SARS-CoV-2 vaccination were the weakest in the liver transplant recipients following a standard vaccine regimen, likely due to their immunosuppression. Interestingly, persons with CLD had a similar neutralization capacity and preserved T-cell–mediated immune response to wild-type SARS-CoV-2 in addition to a similar neutralization capacity of the B.1.1.529 Omicron variant when compared with healthy controls after receiving a standard vaccine regimen and booster vaccination.

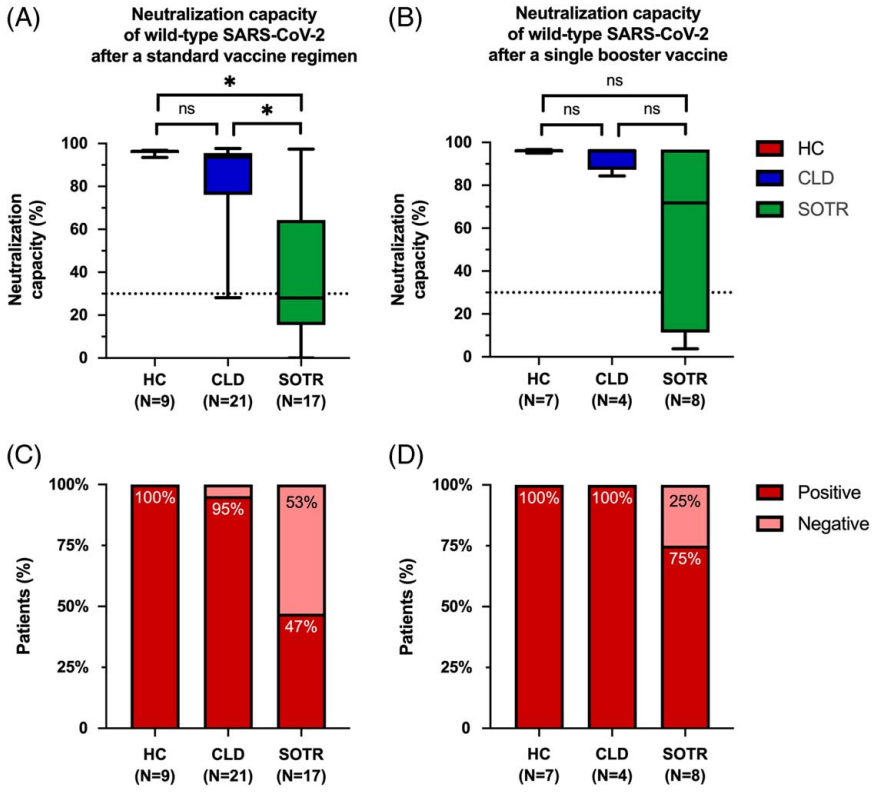

FIGURE 2 Evaluation of neutralization capacity of wild-type SARS-CoV-2 after completion of a standard vaccine regimen (A and C) and following a booster vaccine (B and D) in each group. The boxplots represent the median and interquartile range with whiskers showing the minimum and maximum values. Dotted horizontal lines indicate the cutoff values for positive and negative responses. The respective proportions are provided as bar graphs. Abbreviations: CLD, chronic liver disease; HC, healthy control; SOTR, solid organ transplant recipient; IS, immunosuppressed; WT, wild-type. *$p < 0.05$.

Emerging data suggest that SARS-CoV-2 vaccination generates a variable humoral immune response among persons with CLD. The majority of these studies suggest a similar humoral immune response in persons with CLD compared with healthy controls[6,11,12;] however, Willuweit et al. reported significantly lower antibody titers.[8] Data from the present study demonstrate numerically lower, but not statistically significant differences in anti-S1 protein antibody titers following a standard vaccine regimen in the patients with CLD compared with controls. A larger sample size may be needed to determine if a true response difference is present. Additionally, the emerging literature on SARS-CoV-2 immunity in patients with CLD showed that the presence of cirrhosis did not affect the outcomes or vary by Childs-Pugh class, which is also supported by the findings in this study.[6,8,12] However, Willuweit et al. demonstrated that patients with cirrhosis exhibited a faster deterioration of their antibodies overtime.[8]

Regarding the solid organ transplant recipients, data from the present study demonstrate significantly lower antibody titers and neutralization capacity of the wild-type SARS-CoV-2 in liver transplant recipients when compared with controls following a standard vaccine regimen. Individual tests are important in evaluating specific immune responses, and the evaluation of seropositivity across all the 3 tests may better describe the overall immune response in specific patient populations. Only 20% of SOTRs elicited a positive immune response to the wild-type SARS-CoV-2 in all of the 3 tests following a standard vaccine regimen compared with 75% of controls and 60% of patients with CLD. Our data are consistent with prior studies demonstrating a weaker immune response to SARS-CoV-2 vaccination in this population, especially in older persons and those receiving higher doses of immunosuppressive therapy.[6,7,9,10,13]

Our study was an exploratory evaluation of the T-cell–mediated response against the wild-type SARS-CoV-2 in combination with humoral antibody responses to the wild-type SARS-CoV-2. T-cell–mediated immune response between the patients with CLD and controls was similar yet significantly lower in SOTR when compared with the controls. Clinically, this is important as recent literature suggests that T-cell–mediated immune response may provide protective benefits from SARS-CoV-2 infection by limiting viral replication, improving viral clearance, and supporting long-term immune memory, which can persist despite a weak or undetectable humoral response.[12,16] Furthermore, although variants can partially evade humoral immunity, the T-cell–mediated immunity to these variants appears to be retained.[16–18] Taken together, evaluation of only

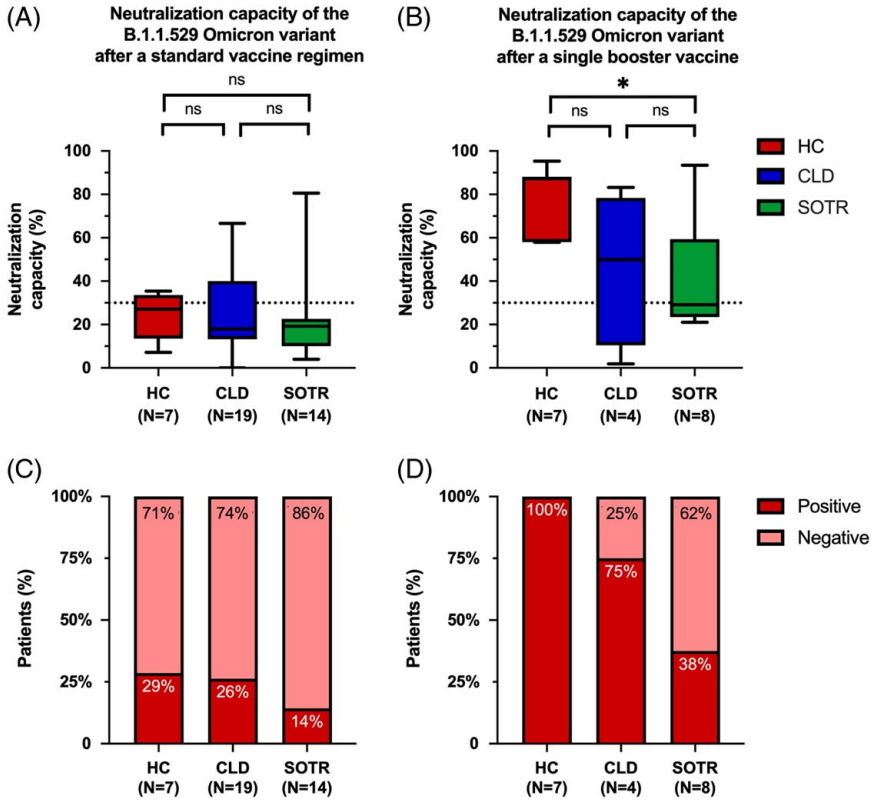

FIGURE 3 Evaluation of neutralization capacity of the B.1.1.529 Omicron variant of SARS-CoV-2 after completion of a standard vaccine regimen (A and C) and following a booster vaccine (B and D) in each group. The boxplots represent the median and interquartile range with whiskers showing the minimum and maximum values. Dotted horizontal lines indicate the cutoff values for positive and negative responses. The respective proportions are provided as bar graphs. Abbreviations: CLD, chronic liver disease; HC, healthy controls; SOTR, solid organ transplant

humoral response may be inadequate to fully characterize the immunity against SARS-CoV-2. Further evaluation of the T-cell–mediated response to SARS-CoV-2 Omicron variants compared to the wild type is needed and will be evaluated in the longer-term longitudinal evaluations with larger sample sizes.

Booster vaccines have been strongly recommended following a standard vaccine regimen, and recent literature suggests that booster vaccines improve the anti-S1 antibody response in CLD and SOTR.[9,13,14] Paired analyses were unable to be performed in patients with CLD and SOTRs due to the small sample size ($N < 5$), but the available data from SOTRs demonstrated that the positive antibody response rose from 41% to 75% and the positive wild-type SARS-CoV-2 neutralization capacity rose from 47% to 75% following a single booster vaccine. Despite detectable antibodies after the booster, concern remains that there may be a lower

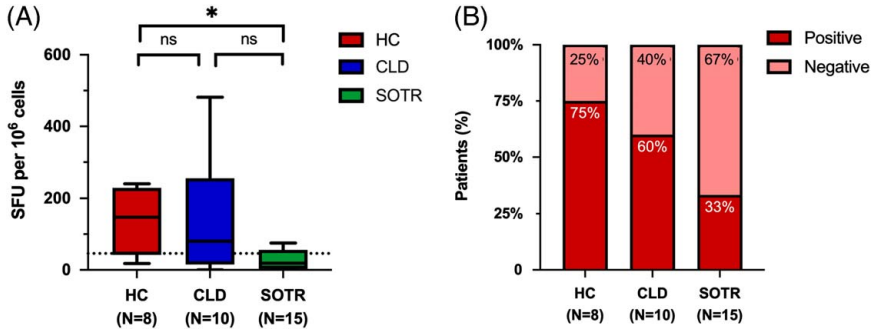

FIGURE 4 Evaluation of T-cell–mediated immune response with ELISPOT to wild-type SARS-CoV-2 after completion of a standard vaccine regimen in each group. The boxplots represent the median and interquartile range with whiskers showing the minimum and maximum values. Dotted horizontal lines indicate the cutoff values for positive and negative responses. The respective proportions are provided as bar graphs. Abbreviations: CLD, chronic liver disease; HC, healthy controls; SOTR, solid organ transplant recipients; SFU, spot forming units. *$p < 0.05$.

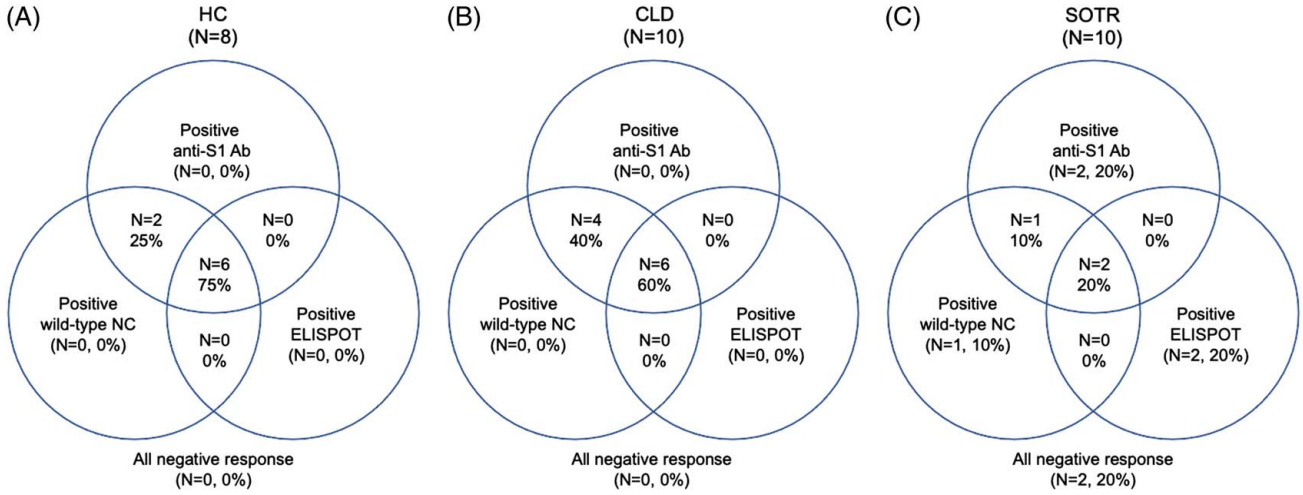

FIGURE 5 Evaluation of seropositivity for anti-S1 antibody, neutralizing capacity, and T-cell–mediated ELISPOT response to wild-type SARS-CoV-2 after completion of a standard vaccine regimen in each group with overlap shown in the Venn diagram. Abbreviations: HC, healthy controls; CLD, chronic liver disease; N, number; SOTR, solid organ transplant recipients.

neutralization capacity for the emerging SARS-CoV-2 variants. These variants acquire mutations in key epitopes in the spike protein for SARS-CoV-2 neutralizing antibodies, which may contribute to immune evasion and reduced vaccine effectiveness against emerging variants.[19] In the general population, Gruell et al.[19] found that receiving a booster significantly improved the serum neutralization of Omicron compared with the 2 doses of the Pfizer vaccine but it was still lower than that of the wild-type SARS-CoV-2. Paired data from our cohort of healthy controls following a standard vaccine regimen and a single, monovalent booster vaccine demonstrated a significantly higher neutralizing capacity of the B.1.1.529 Omicron variant. In addition, 2 recent studies reported a lower B.1.1.529 Omicron-specific neutralization capacity compared with the wild-type SARS-CoV-2 in SOTRs with 3 doses of mRNA vaccine, and many of these patients with a positive anti-RBD response had undetectable levels of Omicron-specific neutralizing antibodies.[3,4] In the present study, the neutralization capacity of the B.1.1.529 Omicron variant following a booster vaccine was significantly lower in SOTR when compared with controls and was similar between CLD patients and controls. Interestingly, the positive neutralization response to the B.1.1.529 Omicron variant in patients with CLD increased from 26% after a standard vaccine regimen to 75% following a single booster and from 14% to 38% in SOTRs, respectively. This has important clinical implications, as neutralization capacity was shown to be predictive of immune protection from infection with the variants of SARS-CoV-2 and could help assess the vaccine efficacy.[20] However, the rapidly evolving mutations in the Omicron variant spike protein limit the generalizability of these findings from Omicron B.1.1.529 to the current Omicron subvariants.[21,22]

Our study evaluated both humoral and T-cell–mediated immune responses to SARS-CoV-2 vaccination in persons with CLD and SOTR patients. A major strength of this study is the use of multiple assays to quantify both humoral and T-cell–mediated immune responses after a standard vaccine regimen (2 doses of BNT162b2 or mRNA-12732). Another strength is the evaluation of the neutralization capacity of the B.1.1.529 Omicron variant in addition to the wild-type SARS-CoV-2 following a standard vaccine regimen and booster vaccination. The study limitations include a relatively small sample size and an observational design that lacks strictly defined intervals between the collection of blood samples for our study cohort. It is possible that larger data sets would discern a statistically meaningful decrement in response rates among those with CLD compared to healthy controls. A methodological strength of this study compared to others is the use of a WHO-standardized measure of vaccine response. Many published studies use semiquantitative, nonstandardized antibody measures. The WHO BAU is an international standard for SARS-CoV-2 Ig and was used in our study to mitigate this limitation. Therefore, our data can be compared to other populations that use similar externally validated standards.

Although immune responses can be quantified, translating these findings into exact clinical implications remains challenging. The correlation between the varied humoral and T-cell–mediated immune responses and the clinical outcomes of COVID-19 in patients with CLD, such as susceptibility to viral infection and severity of disease, is complex and likely multifactorial. This is in part due to the heterogeneity of CLD, which encompasses a broad spectrum of diseases with varying degrees of severity and

immune dysregulation.[1,23] In the immunosuppressed solid organ transplant recipients, the immune response to vaccination is weaker overall. The use of variant-specific booster vaccines or higher vaccine doses may be recommended in these populations to enhance the immunogenicity, but poor CMI responses remain a concern. Alternately, postvaccination testing for both types of immune responses may be needed to determine selective personalized vaccination strategies for individual patients. This is particularly true in an evolving pandemic. Recent data suggest that monoclonal antibody therapy (Evusheld®, or tixagevimab and cilgavimab) frequently utilized in transplant patients may not effectively neutralize certain Omicron variants.[24,25] In addition, new bivalent mRNA booster vaccines have been shown to elicit a stronger and broader immune response to Omicron subvariants compared to the wild-type monovalent booster vaccines.[26] We plan to study the immunologic responses to these agents in future work. Therefore, we conclude that the ongoing longitudinal evaluation of SARS-CoV-2 humoral and cellular responses is valuable and necessary to allow the frequent re-evaluation of an ever-changing viral landscape in immunocompromised individuals.

CONFLICT OF INTEREST

Kenneth Sherman received grants from AbbVie, Gilead, Intercept, and Abbott. The remaining authors have no conflicts to report.

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