The antibody response to COVID-19 among kidney transplant recipients who had PCR confirmed infection

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April 05, 2024

Abstract

Introduction Data on antibody response following COVID-19 in kidney transplant recipients is scarce. We performed a cross-sectional study to investigate antibody response to COVID-19 among kidney transplant recipients. Design We recruited 46 kidney transplant recipients with RT-PCR confirmed COVID-19 and 45 recipients without COVID-19 history. We also constructed two control groups (COVID-19 positive and negative) from a historical cohort of health care workers. We used age and sex-based propensity score matching to select eligible subjects to control groups. We measured SARS-Cov-2 IgG levels quantitatively using the Abbott ARCHITECT system. An antibody level above 1.4 S/C defined positivity. Results Transplant recipients with COVID-19 had a higher BMI, and COVID-19 history in a household member was more common than that of the transplant recipient without COVID-19. IgG seropositivity rate (69.6% vs 78.3%, p=0.238) and median IgG level (3.28 [IQR 0.80-5.85] vs 4.59 [1.61-6.06], p=0.499) were similar in COVID-19 positive transplant recipients and controls. There was a trend toward lower antibody levels in kidney transplant recipients associated with a longer duration between RT-PCR and antibody testing (r=-0.532, p<0.001). Conclusion At the early post-COVID-19 period, transplant recipients have an antibody response that is similar to controls. However, antibody levels and associated immunity should be closely observed with longer follow-up durations.

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eligible subjects to control groups. We measured SARS-Cov-2 IgG levels quantitatively using the Abbott ARCHITECT system. An antibody level above 1.4 S/C defined positivity.

Results

Transplant recipients with COVID-19 had a higher BMI, and COVID-19 history in a household member was more common than that of the transplant recipient without COVID-19. IgG seropositivity rate (69.6% vs 78.3%, p=0.238) and median IgG level (3.28 [IQR 0.80-5.85] vs 4.59 [1.61-6.06], p=0.499) were similar in COVID-19 positive transplant recipients and controls. There was a trend toward lower antibody levels in kidney transplant recipients associated with a longer duration between RT-PCR and antibody testing (r=-0.532, p<0.001).

Conclusion

At the early post-COVID-19 period, transplant recipients have an antibody response that is similar to controls. However, antibody levels and associated immunity should be closely observed with longer follow-up durations.

What's already known about this topic?

Data on the seropositivity rate following COVID-19 in kidney transplant recipients are scarce. Recent studies have reported antibody response rates ranging between 41.0-80.0% in kidney transplant recipients following recovery from COVID-19 infection.

What does this article add?

We found post infectious antibody response similar to general population. However time dependent decrease of antibody levels should be followed.

Keywords: COVID-19; immune response; immunosuppression; kidney transplantation; SARS-CoV-2 IgG antibody

Introduction

Coronavirus disease 2019 (COVID-19) pandemic caused by SARS-CoV-2 impacted the global health system in an unprecedented way. The rapid development of various vaccines has become an important milestone in combating the pandemic. Vaccination relies on the production of antibodies via stimulation of humoral immunity. However, data regarding antibody response to infection in immunosuppressed patients, including kidney transplant recipients, is not well defined.

Different mortality rates for COVID-19 were reported from different countries. According to a recent study in western countries, mortality rates changed between 4.0% and 16.1% for reported cases. Higher mortality rates were reported in patients with kidney disease, specifically, in patients with kidney transplantation mortality ranging between 18.0- 41.6% was reported in different studies [1-6]. Moreover, data on the seropositivity rate following COVID-19 in kidney transplant recipients are scarce and not in agreement across studies. According to a recent study, lower antibody response rates (41.0%) were reported in kidney transplant recipients following recovery from the infection [7,8]. On the other hand, Azzi et al. investigated 69 kidney transplant recipients with reverse transcriptase-polymerase chain reaction (RT-PCR) confirmed COVID-19 infection, and they found anti-nucleocapsid antibodies in 55 (80.0%) of them [9].

Usually, higher antibody response rates were reported for patients from the general population [8]. In a recent study, anti-nucleoprotein seropositivity was found as 78.2% among health care workers with RT-PCR confirmed COVID-19 [10].

In this cross-sectional study, we aimed first to investigate the prevalence of anti–SARS-CoV-2 antibodies in kidney transplant recipients; secondly, we examined the factors associated with the absence of antibody response.

Material and Methods

Study design

We performed a cross-sectional study to recruit renal transplant recipients who were under regular follow-up in two university hospital's transplantation centers (CMF and KMF) and had RT-PCR confirmed COVID-19. We also recruited consecutive renal transplant recipients who did not have a history of COVID-19 and were attending transplantation outpatient clinics in those institutions. We constructed two additional control groups from a previously screened cohort of health care workers [10]. None of the included patients were vaccinated for COVID-19 prior to antibody measurement.

We used descriptive comparative design to assess the outcomes. The study protocol was approved by the local medical ethics committee (approval no: 2021-2921) and the Ministry of Health's Scientific Committee (approval no: 2020-11-30T14_57_30). The study was conducted in accordance with the 1975 Declaration of Helsinki, as revised in 2013.

Sampling

The study was conducted between December 7, 2020, and February 12, 2021. The date of blood specimen collection for antibody measurement was accepted as the enrollment date to the study. The date of RT-PCR testing was accepted as the first day of infection.

Transplant recipients

Based on previous studies we predicted seropositivity following COVID-19 as 60% for transplant recipients [9,11] and 90% for subjects from the general population [12-14]. We performed a power analysis, and we planned to recruit 42 participants to each group.

A total of 623 patients were attending outpatient transplantation clinics during the year 2020. Target population was patients who had COVID-19 following April, 2020. We did not formally screen all patients under follow-up; however, all of our transplant patients who had an RT-PCR confirmed COVID-19 history were eligible for the study. We located 57 patients who had COVID-19, one of them died before the start of the study, 46 of them accepted to participate in the study.

We recruited transplant patients who gave informed consent to the COVID-19 negative group if they declared that they did not have a diagnosis of COVID-19 as of the recruitment day. We also checked if they had any RT-PCR test due to mandatory screening (before hospitalization due to any cause, having a household member with COVID-19) and confirmed that the RT-PCR test was negative.

Controls

We recruited control subjects from a cohort of health care workers that we examined previously [10]. Details of recruitment and data collection for those participants were previously described in detail [10].

In that cohort, 116 subjects were RT-PCR positive. We excluded any subjects with malignancy or using immunosuppressive drugs. We transformed the duration between RT-PCR and antibody testing to binomial data based on eight weeks' cut-off value. RT-PCR positive control group is formed by recruiting subjects using propensity score matching based on age, sex, and transformed antibody testing duration data with a 1:1 ratio.

Among healthcare workers who do not have COVID-19 history, we selected subjects designated as "no risk" (health care workers who were not attending the hospital because of administrative changes related to pandemic) regarding COVID-19. We excluded any subjects with malignancy or using immunosuppressive drugs; 106 subjects were eligible for selection. We used age and sex-based propensity score matching to select subjects from this cohort with a ratio of 1:1.

We used the same laboratory procedures to measure antibody levels in those subjects and transplant recipients.

Data Collection

We filled a standard form for every patient, and we used patient interviews, medical records of the patients, the hospital's electronic database, and the national public health data management system to collect data. Our form consisted of the following parts; demographics, clinical data including transplantation history, drug use, laboratory parameters, history, and clinical data related to COVID-19, and computed tomography (CT) findings. We also used the COVID-19 severity index to classify the patients into five mutually exclusive categories; asymptomatic or presymptomatic, mild, moderate, severe, critical illness [15].

PCR testing and Assessment of Antibodies

We used the same methods for RT-PCR testing and SARS-CoV-2- antibody measurement as described in detail previously [10,16]. For detection of COVID-19 RNA, a commercial RT-PCR kit (Bio-Speedy COVID-19 RT-qPCR kit; Bioeksen R & D Technologies Ltd., Istanbul, Turkey) was used. For detection of SARS-CoV-2 IgG (anti-nucleocapsid protein antibodies), chemiluminescent microparticle immunoassay (Abbott Laboratories, Cat no: 6R86, Lot no: 16253FN00) was carried out according to the manufacturer's instructions, and samples were run on the related instrument (ARCHITECT, Abbott Laboratories, Abbott Park, IL, USA). Qualitative results were reported by the instrument with the cut-off value of 1.40 S/C as recommended.

Statistical Analysis

Descriptive data were presented as mean and ± standard deviation (SD) and median and interquartile range (IQR) for the continuous variables and frequency and percentages (%) for the categorical variables. Continuous variables were evaluated for normality distribution using Shapiro-Wilk test. Kidney transplant recipients and control groups were compared with an independent sample t test for normally distributed variables and Mann-Whitney U test for non-normally distributed variables. Categorical variables were compared by using Chi Square or Fisher's Exact test for proportion. Multivariate analysis was applied to determine association between antibody level, group and post-infection duration. All significance tests were 2-tailed, and values of $p < 0.05$ were considered statistically significant. All statistical analyzes were performed by SPSS software version 21 (Chicago, IL).

We employed propensity score matching to balance in observed baseline covariates and reduce the bias of treatment effect between kidney transplant recipients and control groups. We assumed at ratio of 1:1 on age and sex with nearest neighbor matching method. The propensity score matching was performed using the RStudio (version 4.0.2 software).

According to previous studies, we accepted a seropositivity rate following COVID-19 as 60% for transplant patients and 90% for the general population. Therefore, considering the percentage of previous studies, we performed power analysis (Gpower software, version 3.1, Kiel, Germany) with 90% power and an error of 0.05 to determine the minimum sample size for RT-PCR positive kidney transplant recipients and control groups. A minimum sample size of 42 was estimated in each group.

Results

Demographic, Clinical and Laboratory data

We recruited a total of 91 kidney transplant recipients, 46 of them had an RT-PCR confirmed COVID-19, and 45 of them did not have COVID-19 history (Table 1). Demographic, clinical, and laboratory data of transplant recipients grouped according to COVID-19 status are shown in Table 1. Both groups were similar regarding age and sex. Etiology of CKD and donor type and post-transplant duration were also similar between the two groups. However, patients with COVID-19 had a higher BMI, and COVID-19 history in a household member was more common among them. Other parameters were similar between the two groups.

The majority (95.6%) of the patients with COVID-19 were symptomatic, and according to thorax computed tomography examination, 30 (65.2%) patients had findings compatible with COVID-19. There was no need for hospitalization in 12 (26.1%) patients; the remaining 34 (73.9%) patients were hospitalized, 16 (34.8%) patients needed oxygen, and three (6.5%) of them were followed up in the intensive care unit. Two (4.3%) patients needed intubation. Except for one patient who died on the 26th day of the infection, all patients recovered from COVID-19. The hospitalization duration was 11.7±7.9 days (median:9 days, range:3-38 days). According to the COVID-19 severity index, two (4.3%) patients were asymptomatic or presymptomatic, 15 (32.6%) patients had a mild illness, 16 (34.7%) patients had a moderate illness, 10 (21.7%) patients had a severe illness, and three (6.5%) patients had a critical illness.

Seropositivity

Seropositivity rate and IgG levels among kidney transplant recipients and controls stratified by COVID-19 status are shown in Table 2. Among subjects with COVID-19 history, the SARS-Cov-2 IgG positivity rate and IgG level of kidney transplant recipients were similar to that of the control group. The frequency of COVID-19 related symptoms was more common among kidney transplant recipients than that of the controls; however, the frequency of pulmonary involvement assessed by computerized tomography was similar between the two groups. There was no statistically significant difference between the kidney transplant recipients and controls in terms of the duration from RT-PCR to antibody testing. (Table 2).

Among subjects without COVID-19 history, three kidney transplant recipients had positive IgG antibodies, and two of them had a history of COVID-19 in a household member. SARS-Cov-2 IgG antibody positivity rate and IgG level of kidney transplant recipients were similar to that of the control group (Table 2).

Predictors of antibody positivity

We compared demographic, clinical, laboratory, and treatment-related data of the transplant patients who developed antibodies with those who did not (Table 3). The median duration between RT-PCR and antibody testing was shorter (37.5 [IQR 20.5-57.8] vs 82.5 [IQR 52.3-105.0] days, p=0.01) in patients who had SARS-Cov-2 IgG antibodies compared to that of the patients who do not have IgG antibodies. There were no statistically significant differences among the two groups regarding demographic, clinical, laboratory parameters. Additionally, the cessation rate of different immunosuppressive drugs was also similar between the two groups.

As an additional analysis, we looked at the correlation between different laboratory parameters (peak CRP, peak ferritin, fibrinogen, peak d-dimer, peak procalcitonin, e-GFR) and level of SARS-Cov-2 IgG antibodies. There was no significant correlation between those parameters (data not shown).

Finally, we analyzed the correlation between SARS-Cov-2 IgG antibody levels and the duration between RT-PCR and antibody testing. The antibody level in kidney transplant recipients and controls according to duration following RT-PCR testing is shown in Figure 1. Visual examination revealed that there was a trend toward lower antibody levels in kidney transplant recipients with increasing post-infection duration.

There was a significant correlation between antibody level and duration in transplant recipients (r=-0.532, p<0.001), however, there was no statistically significant correlation between antibody level and duration in controls (r=0.198, p=0.186). Additionally, we constructed a multivariate regression model; we used antibody levels as the dependent variable, and the study group along with the duration following RT-PCR testing as the independent variables. This analysis showed that the study group was not an independent determinant of antibody levels (data not shown).

Discussion

We found that kidney transplant recipients developed an antibody response following COVID-19; the mean antibody level and the seropositivity rate were similar to that of the control group. To the best of our knowledge, the largest report about antibody response in kidney transplant recipients was from Azzi et al. [9]. They examined 69 kidney transplant recipients who had an RT-PCR confirmed COVID-19 diagnosis, and 55 (80.0%) of them have positive antibody response. They used the same test as our study to measure antibody levels. They measured the antibody response following a median of 44 days following RT-PCR positivity. Hartzell et al. examined anti-SARS-Cov2 IgG antibodies in 16 kidney transplant recipients following a mean of 16.1 days of RT-PCR testing; antibody positivity rate was reported as 60.0% and 63.6% depending on immunosuppressive drug use [17]. Burack et al. examined 39 kidney transplant recipients and found an antibody positivity of 41.0% [7].

According to our data infection in a household member and high BMI was associated with COVID-19 infection in kidney transplant recipients. Previous studies showed an association between high BMI and severe COVID-19 [18]. However, it is not clear whether the risk of COVID-19 increases with high BMI [19-22]. A recent study showed that the prevalence of obesity in patients hospitalized with COVID-19 is higher than the worldwide prevalence of obesity [23].

In kidney transplant recipients who did not have positive RT-PCR testing, we found a similar SARS-CoV-2 IgG antibody positivity rate to our controls. In line with our results, a recent study reported that the seroprevalence rate was 6.6% in asymptomatic people [24]. In another study SARS-CoV-2 seroprevalence in healthy blood donors was reported as 3% [25].

We did not identify any specific risk factor for lack of seroconversion following COVID-19. However, we noted a trend toward lower antibody levels in patients who had a longer post-infection duration. A similar observation was reported by Benotmane et al. [26]. They examined 29 kidney transplant recipients hospitalized for COVID-19 and measured antibody levels up to 6 months after COVID-19. During the follow-up, 20.7% of the patients become seronegative. A considerable IgG reduction was observed in patients treated with calcineurin inhibitors and steroids. No statistically significant difference was found regarding disease severity.

Our study has some limitations; we did not formally screen all patients; we might have overlooked some patients who had severe COVID-19 and died. However, it is unlikely that we missed mild cases because most of the patients were in close telephone contact with transplant coordinators during this period. Another limitation of this study is that the date of infection was defined as the date of PCR positivity, as opposed to the date of symptom onset. Since transplant recipients may exhibit prolonged shedding of the virus, the date of PCR positivity may not always be an accurate estimate of infection onset. Kidney transplant recipients and controls had different distribution characteristics regarding duration between COVID-19 and antibody testing, all the controls were tested for antibodies following at least four weeks, and no control was tested following more than 12 weeks. However, the distribution of transplant recipients between different IgG testing duration categories was homogenous. Finally, we did not examine parameters related to cellular immunity.

Conclusions

Kidney transplant recipients seem to have an antibody response similar to the general population at the early post-COVID-19 period. However, similar to the general population there is a tendency toward lower antibody levels with increasing post-infection duration. Therefore, we suggest caution for humoral immunity in kidney transplant recipients following COVID-19; at least following three months post-infection; follow-up of antibody levels and booster vaccination might be warranted.

Author contributions

The individual contribution of each co-author is listed below using the following keys: 1-Conception or design, or analysis and interpretation of data, or both, 2-Drafting the article or revising it, 3-Providing intellectual content of critical importance to the work described. 4-Final approval of the version to be published.

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Table 1 . Demographic, clinical, and laboratory parameters of kidney transplant recipients grouped according to COVID-19 status.

	COVID-19 (+) n=46	COVID-19 (-) n=45	p value
Age (years)	46.5 (36.8-55.0)	37.0 (32.0-55.0)	0.194
Male, n (%)	32 (69.6)	27 (60.0)	0.231
Etiology of CKD	Etiology of CKD	Etiology of CKD	Etiology of CKD
Diabetes Mellitus, n (%)	5 (10.9)	3 (6.7)	0.599
Glomerulonephritis, n (%)	8 (17.4)	11 (24.4)	
Others, n (%)	33 (71.7)	31 (68.9)	
Living donor, n (%)	36 (78.3)	35 (77.8)	0.600
Transplantation Duration (months)	108.0 (48.0-147.0)	84.0 (24.0-132.0)	0.378
COVID-19 in a household member, n (%)	29 (64.4)	3 (6.8)	<0.001
Comorbidities	Comorbidities	Comorbidities	Comorbidities
Diabetes, n (%)	13 (28.3)	11 (24.4)	0.431
Hypertension, n (%)	31 (67.4)	26 (57.8)	0.232
BMI (kg/m^2)	27.4 ± 4.5	24.7 ± 4.2	0.004
Serum Creatinine (mg/dL)	1.28 (1.01-1.53)	1.23 (1.02-1.72)	0.778
e-GFR+ ($ml/min/1.73 m^2$)	64.9 ± 24.1	64.6 ± 31.6	0.955
Baseline immunosuppression	Baseline immunosuppression	Baseline immunosuppression	Baseline immunosuppression
Steroids, n (%)	46 (100.0)	42 (93.3)	0.117
Calcineurin inhibitor, n (%)	43 (93.5)	43 (95.6)	0.511
Mycophenolic acid derivates, n (%)	41 (89.1)	35 (77.8)	0.119
m-TOR inhibitors, n (%)	4 (8.7)	4 (8.9)	0.631
Azathioprine, n (%)	3 (6.5)	7 (15.6)	0.149

Values are presented as mean ± SD and median with IQR.

CKD: Chronic kidney disease, BMI: Body mass index, m-TOR inhibitors: Mechanistic target of rapamycin inhibitors.

+Calculated using CKD-EPI formula.

Table 2 . Seropositivity among kidney transplant recipients and controls stratified by COVID-19 status.

	COVID-19 (+) Kidney Transplant recipients n=46	COVID-19 (+) Controls n=46	p value	COVID19 (-) Kidney Transplant recipier
Age (years)	45.9 ± 12.1	41.3 ± 10.2	0.053	37.0 (32.0-55.0)
Male, n (%)	32 (69.6)	32 (69.6)	0.589	27 (60.0)

Symptoms, n (%)	44 (95.7)	36 (78.3)	0.013	NA
CT Result, n (%)	30 (65.2)	30 (65.2)	0.558	NA
IgG positivity rate, n (%)	32 (69.6)	36 (78.3)	0.238	3 (6.7)
IgG level (S/C)	3.28 (0.80-5.85)	4.59 (1.61-6.06)	0.499	0.03 (0.02-0.05)
Days following RT-PCR test	49.5 (25.8-70.6)	55.0 (49.3-61.0)	0.392	NA

Values are presented as mean ± SD and median with IQR.

CT: Computed tomography Ig: Immunoglobulin RT-PCR: Reverse transcriptase-polymerase chain reaction.

Table 3. Comparison of transplant recipients with and without IgG following RT-PCR confirmed COVID-19

	IgG positive n=32	IgG negative n=14	p value
Age (years)	47.5 ± 11.1	42.2 ± 13.8	0.172
Male, n (%)	23 (71.9)	9 (64.3)	0.427
Etiology of CKD	Etiology of CKD	Etiology of CKD	Etiology of CKD
Diabetes Mellitus, n (%)	4 (12.5)	1 (7.1)	0.364
Glomerulonephritis, n (%)	7 (21.9)	1 (7.1)	
Others, n (%)	21 (65.6)	12 (85.7)	
Living donor, n (%)	24 (75.0)	11 (84.6)	0.392
Transplantation Duration (months)	108.9 ± 61.9	88.0 ± 78.8	0.337
Comorbidities	Comorbidities	Comorbidities	Comorbidities
Diabetes, n (%)	10 (31.3)	3 (21.4)	0.381
Hypertension, n (%)	22 (68.8)	9 (64.3)	0.511
Coronary artery disease, n (%)	7 (21.9)	5 (35.7)	0.264
Chronic obstructive pulmonary disease, n (%)	2 (6.3)	1 (7.1)	0.673
BMI (kg/m^2)	27.6 ± 4.3	26.9 ± 5.1	0.529
Serum creatinine (mg/dL)	1.33 (1.03-1.48)	1.15 (0.87-1.83)	0.867
e-GFR+(ml/min/1.73 m^2)	63.5 ± 19.0	68.3 ± 33.6	0.537
Disease severity index	Disease severity index	Disease severity index	Disease severity index
Asymptomatic to mild, n (%)	21 (66.0)	12 (85.0)	0.286
Severe to critical, n (%)	11 (34.0)	2 (15.0)	
Days following RT-PCR test	37.5 (20.5-57.8)	82.5 (52.3-105.0)	0.001
Lowest white blood cell count, (/mm^3)	4925.7 ± 1748.0	4596.8 ± 1667.2	0.582
Lowest lymphocytes count, (/mm^3)	791.0 ± 603.0	665.4 ± 314.7	0.875
Peak CRP, (mg/dL)	75.8 ± 66.5	74.7 ± 58.0	0.813
Peak ferritin, (ng/mL)	625.0 (249.5-1468.3)	670.1 (236.0-1245.0)	0.839
Fibrinogen, (mg/dL)	447.8 (4.98-649.8)	329.1 (4.76-515.5)	0.439
Peak D- Dimer, (μg/mL)	1.03 (0.36-2.94)	0.91 (0.66-2.78)	0.868

Peak procalcitonin, (ng/mL)	1.72 (0.92-127.0)	92.5 (1.13-191.3)	0.146
Peak uric acid, (mg/dL)	8.1 ± 2.1	7.7 ± 2.3	0.622
Stopping MPA, n (%)	17 (53.1)	7 (50.0)	0.549
Stopping CNI, n (%)	3 (9.4)	0 (0.0)	NA
Stopping MPA or CNI, n (%)	18 (56.3)	7 (50.0)	0.471

Values are presented as mean ± SD and median with IQR.

CKD: Chronic kidney disease, BMI: Body mass index, RT-PCR: Reverse transcriptase-polymerase chain reaction MPA: Mycophenolic acid derivates, CNI: Calcineurin inhibitor

+Calculated using CKD-EPI formula.

Figure Legends

Figure 1. SARS-CoV-2 IgG antibody level among transplant recipients and controls, according to duration between RT-PCR and IgG testing.