COVID-19 outbreak in vaccinated patients from a hemodialysis unit:

antibody titers as a marker of protection from infection

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Running title: COVID-19 outbreak after BNT162b2 mRNA vaccination

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ABSTRACT

Background. Patients on maintenance hemodialysis have an increased risk of severe COVID-19 and a reduced response to vaccines. Data are needed to identify immune correlates of protection in this population.

Methods. Following a COVID-19 outbreak among vaccinated patients in a hemodialysis unit, clinical data and serological response to BNT162b2 vaccine were retrospectively recorded.

Results. Among fifty-three patients present in the dialysis room, fourteen were infected by SARS-CoV-2 alpha variant (COVID_Pos) and 39 were not. In comparison to uninfected patients, COVID_Pos patients more frequently had additional causes of immunosuppression (50% vs 21%, p=0.046), and were more often scheduled on the Monday-Wednesday-Friday (MWF) shift (86% *vs* 39%, p=0.002). Moreover, COVID_pos had lower anti-Spike IgG titers than uninfected patients (24 BAU/ml [3-1163] vs 435 BAU/mL [99-2555], p=0.001) and lower neutralization titers (108 [17-224] vs 2483 [481-43908], p=0.007). Anti-Spike and neutralization antibody titers are correlated (r=0.92, p<0.001). In multivariable analysis, MWF schedule (OR=10.74 (1.9-93.5), p=0.014) and anti-spike IgG titers one month before the outbreak (<205 BAU/ml: OR=0.046 (0.002-0.29), p=0.006) were independently associated with COVID-19 infection. None of the patients with anti-Spike IgG above 284 BAU/mL got infected. Ten out of fourteen COVID_Pos patients were treated with Casirivimab and Imdevimab. No patient developed severe disease.

Conclusions. Anti-spike IgG titer measured prior to exposure correlates to protection from SARS-CoV-2 infection in hemodialysis patients. BNT162b2 vaccination alone or in combination with monoclonal antibodies prevented severe COVID-19.

Keywords: BNT162b2 mRNA vaccination, COVID-19 outbreak, hemodialysis, humoral response, monoclonal antibodies

INTRODUCTION

Infectious diseases are the second highest cause of death in end-stage renal disease (ESRD) patients.[1] Patients on hemodialysis are vulnerable to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection (coronavirus disease 2019 [COVID-19]), with reported mortality rates over 20%.[2-4] Consequently, patients on dialysis were among the first to be vaccinated against SARS-CoV-2 in France. However, the effectiveness of vaccination is often lower in these patients as compared to healthy subjects.[5, 6] After two injections of BNT162b2, between 79 and 90% of hemodialysis patients seroconvert, a rate above kidney transplant recipient's, but below the general population's.[7-11] Moreover, real-world data on vaccine efficacy or immune correlates of protection are not yet available for this particular population. Thus, the retrospective investigation of breakthrough infections in vaccinated dialyzed patients is of critical importance to optimize prophylactic and therapeutic interventions in ESRD patients.

Here, we described an epidemic cluster of SARS-CoV-2 alpha variant in our dialysis unit, two months after the end of the vaccination campaign. We report patient's outcomes, provide our feedback on the epidemic management, and identify a correlate of protection for this outbreak.

MATERIALS AND METHODS

Cohort and context

We performed a retrospective mono-centric study in the hemodialysis unit of Necker Hospital, Paris, France, following an outbreak of SARS-CoV-2 alpha variant infections among patients.

In April 2021, 53 patients, rotating in two shifts, either Monday-Wednesday-Friday (MWF) or Tuesday-Thursday-Saturday (TTS), were undergoing hemodialysis in our unit. The dialysis unit is a single room containing 14 hemodialysis stations (**Figure 1**). The distance between each station is of at least two meters. Since March 2020, protective measures are taken to limit COVID-19 spread. Patients and caregivers are required to wear chirurgical masks. A check-in was set up at the entrance to welcome patients one-by-one, sanitize hands, take the temperature with a temperature gun and ask about their epidemiologic contact history and state of health with a focus on fever, cough, dyspnea, rhinorrhea and diarrhea. Patients do not share transport facilities.

Virological parameters

SARS-CoV-2 infections during the outbreak were confirmed through real-time RT-PCR assay of nasopharyngeal swab specimens. Viral loads, estimated through the Ct at diagnosis, were classified as very low (Ct>35), low (30-34), moderate (25-29), and high (<25).

Clinical parameters

At the time of the study, patients were considered vaccinated if they had received either two injections of vaccine or one injection following a SARS-CoV-2 infection. Patients were considered to have a history of SARS-CoV-2 infection if they had had a positive PCR on

nasopharyngeal swab or positive anti-nucleocapsid (N) serology prior to vaccination. All vaccinated patients received the BNT162b2 mRNA vaccine. Mild, moderate and severe forms of COVID-19 were defined as follow: no oxygen, <4 L per min, or ≥ 4L per min, respectively. Lymphopenia was defined as a lymphocyte count <1 G/L.

Sampling

Humoral response against SARS-CoV-2 was assessed on sera collected before vaccination (referred to as M-3), one month after the second vaccine dose (M-1), and at the beginning of the outbreak (M0). The outbreak occurred one month after the M-1 timepoint.

Serological assays

Anti-SARS-CoV-2 IgG antibodies directed against the spike protein and the RBD domain of the S1 subunit were measured using CMIA (chemiluminescent microparticle immunoassay) with the IgG II Quant anti-SARS CoV-2 (Abbott, Issy les Moulineaux) (M-1: n=42, M0: n=43). Results ≥7.1 BAU/mL (Binding Antibody Units), equivalent to ≥50 AU/mL (Arbitrary units) were considered as positive according to the manufacturer recommendations.[12] SARS-CoV-2 neutralization was assessed using the S-fuse assay, as previously described (M-3 n= 28, M-1 n=35, M0 n=39).[13]

Ethics

Data collection was declared to the French Commission Nationale de l'Informatique et des Libertés. This protocol was approved by our local ethic committee (Comité d'éthique de la recherche AP-HP centre; IRB: #00011928)

Statistical Analyses

Categorical and continuous variables were expressed as count (percentage) and median (interquartile range, IQR), respectively. Fisher's exact tests were used for categorical comparison and Mann–Whitney for continuous variables. Variables associated with COVID-19 infection post-vaccination were analyzed by logistic regression. All variables with a *P* value <0.2 in univariable analysis were included in the multivariable analysis. Stepwise backward selection based on the Akaike information criterion was used for the final multivariable model (logistic regression). Results were analyzed with GraphPad Prism v9.0.0 and R v4.0.3.

RESULTS

Description of the outbreak

On Friday April 16th, a patient undergoing hemodialysis in our unit was hospitalized for dyspnea and diagnosed with SARS-CoV-2 alpha variant. She was on the MWF morning hemodialysis shift. Medical, and paramedical staff of the dialysis unit as well as all patients undergoing hemodialysis were tested with naso-pharyngeal PCR swabs on Monday and Tuesday, April 19 and 20, 2021. This testing identified 10 additional cases: four patients on MWF morning shift, three patients on MWF afternoon shift, two patients on TTS afternoon shift and one caregiver of the dialysis unit.

The next week (April 26) four additional cases were identified (three patients on MWF morning shift and one patient on MWF afternoon shift). This weekly basis testing strategy was maintained for two additional weeks, without new cases, and stopped on Tuesday May, 11 (**Figure 2**)

In total, fourteen infections with the Alpha variant were diagnosed (hereafter referred to as the COVID_Pos group, n=14) in the week following the identification of the first case. Among the rest of the 53 hemodialysis patients, 39 remained uninfected (forming the COVID_Neg group, n=39) (**Figure 2**).

Management of the COVID_Pos Patients

At the time of the study, 50 patients were fully vaccinated with a median time between the last BNT162b2 injection and the outbreak of 68 days [55-69]. Among the three patients who refused vaccination, two had a positive PCR test during the outbreak (66.6%, p=0.13). At diagnosis, 6 infections were asymptomatic, 6 were mild and 2 moderate. PCR-based screening identified Alpha SARS-CoV-2 variant in all patients. The viral load was high in nine patients and low or very low in five. All COVID_Pos patients were isolated on a dedicated schedule on MWF morning to stop inter-individual contaminations. Because these patients had risk factors for severe COVID-19 (**Table 1**), and in accordance with the local recommendations, we decided to treat 10 out of the 14 COVID_Pos patients with monoclonals antibodies (mAbs) (Casirivimab (1200mg) and Imdevimab (1200mg) (120 mg/mL, Roche)). Six patients were hospitalized because of COVID-19 (including one patient who refused vaccination), but none required more than 3 L per min of oxygen (**Table 1**). None of the COVID_Pos patients died. Of note, one patient developed hypotension after the mAb infusion, which resolved quickly after crystalloid infusion.

Comparison between infected and non-infected patients

Most of the cases were diagnosed on the MWF shift (86% of the COVID_Pos patients). There was no statistically significant difference between COVID_Pos and COVID_Neg patients

regarding median age (67 *vs* 64 years, p=0.8), sex (39% of female *vs* 57%, p=0.23), cause of ESRD (p=0.19), or prior SARS-CoV-2 infection (14% vs 26%, p=0.5). An additional cause of immunosuppression was more frequently found in patients belonging to the COVID_Pos than COVID_Neg group (50% *vs* 21%, p=0.046) (**Table 2**) and on the MWF schedule than in the TTS schedule (46% vs 11%, p=0.01) (**Table 3**).

Anti-S IgG and neutralization titers of the vaccinated patients were retrospectively measured at the peak of humoral response to vaccination, i.e. one month after the last vaccine dose (M-1). At M-1, 35/42 (83%) patients had a positive serology. Median M-1 anti-S IgG titer was 224 BAU/mL [16-1145] and median M-1 neutralization titers were 1045 [75-7541]. COVID_Pos patients had lower M-1 antibody titers (24 BAU/ml [3-1163] *vs* 435 BAU/mL [99-2555], p=0.001) and lower neutralization titers than the COVID_Neg patients (108 [17-224] vs 2483 [481-43908], p=0.007) (**Figure 3A**). None of the patients with M-1 anti-Spike IgG above 284 BAU/mL got infected by SARS-CoV-2. Neutralization titers and anti-S antibody quantifications were well correlated (r=0.92, p<0.001) (**Figure 3B**). Patients in the MWF schedule tended to display lower anti-S IgG (117 BAU/ml [4-506] vs 386 BAU/ml [110-1837], p=0.06) and lower neutralization titers (174 [14-3261] vs 2107 [636-60632], p=0.06) than patients in the TTS schedule (**Figure 3C**). As expected, individuals who received one dose of vaccine following a SARS-CoV-2 infection exhibited higher levels of antibodies (794 BAU/mL [168-5023] versus 183 [8-788], p =0.09) and neutralizing titers (111422 [10210-260273] versus 371[20-1770], p =0.0005) than naïve individuals who received two doses of vaccine (**Figure 3D**). Conversely, vaccinated patients with an additional cause of immunosuppression had a decreased antibody response (anti-S IgG: 2 BAU/mL [24-150] versus 381 BAU/mL [141-

1915], p =0.004; neutralizing titers: 89 [14-264] versus [1750 175-24201], p =0.03) (**Figure 3E**).

Evolution of neutralization titers overtime showed that in the COVID_Neg group, titers wanned, whereas antibody titers rose in the COVID_Pos group, to reach levels comparable to those of the COVID_Neg group (**Figure 3F**).

In multivariable analysis, MWF schedule (OR=10.74 (1.9-93.5), p=0.014) and M-1 anti-spike (S) antibodies levels (>205 BAU/ml: OR=0.046 (0.002-0.29), p=0.006) were independently associated with COVID-19 infection (**Table 4**).

DISCUSSION

Here, we report an outbreak of SARS-CoV-2 alpha variant in a dialysis unit, two months after the end of the vaccination campaign, showing that neutralizing titers and anti-S IgG levels can be correlated with the risk of infection. Our data further support the protecting effect of vaccination either alone or in combination with mAb therapy in this population.

Identifying immune correlates of protection from SARS-CoV-2 is critical to forecast individual protection and determine if/when a booster dose is needed. Currently, detection of neutralizing antibodies is considered as the best predictive factor of protection against SARS-CoV-2 infection.[14-16] It has been suggested that the peak neutralizing titer (measured in the month following complete vaccination) is a marker of the overall immune response and is more predictive of protection than the peri-infection neutralizing titer.[17] Indeed, in our study, lower peak neutralization titers were significantly associated with the later

occurrence of SARS-CoV-2 infection. Patients of the MWF schedule were more immunocompromised and had lower neutralization titers than patients of the TTS schedule, which might explain why the outbreak spread more rapidly within this group.

Neutralization titers are not readily available in the daily clinical practice, and a more practical immune correlate of protection, such as the anti-S IgG level, would be useful. In our study and others[18], a correlation between anti-S IgG and neutralization titers was observed, suggesting that peak anti-S IgG levels could be used as markers for protection. Accordingly, peak anti-S IgG were significantly lower in the COVID_Pos group in comparison with the COVID_Neg group. Moreover, within the MWF group, the occurrence of a SARS-CoV-2 infection was also associated with lower peak anti-S IgG levels. Altogether, it suggests that the level of antibody obtained one month after a second dose of vaccination is predictive of protection among hemodialysis patients. Of note, no SARS-CoV-2 infection (with the Alpha variant) was observed in patients with peak anti-S IgG titers above 284 BAU/mL, an observation in line with a recent report suggesting a threshold of protection for Alpha of 264 BAU/ml of anti-S IgG in the general population.[19]

Interestingly, the level of neutralization titers rose at the time of SARS-CoV-2 infection to levels comparable to those observed in the COVID_Neg group, hence possibly to protective levels. This observation is in favor of a third vaccine dose in this fragile population, as recommended by the French National Authority for Health.

Whether vaccination protects efficiently against severe COVID-19 infections in dialysis patients is currently unknown. El Karoui et al. modelized the expected number of SARS-CoV-

2 severe infections and identified vaccination coverage in French hemodialysis patients as independently associated with protection against severe infection.[20] Likewise, Espi et al. used the French Renal Epidemiology and Information Network (REIN) to compare 1474 cases of COVID-19 in hemodialysis after none, one or two vaccine doses. They demonstrated a reduction of COVID-19 incidence and severity after vaccination; nevertheless, mortality among the patients who had received two doses still reached 11% .[21] In our cohort, no severe infection occurred. This observation might be because most of the patients were rapidly treated with Casirivimab and Imdevimab. The efficacy of this strategy is, however, difficult to assess as there was no control group in our study. Yet, as dialysis patients are at higher risk of severe infections[2-4] and mAb perfusions appear to be safe, we believe that this a reasonable strategy. Of note, this approach was then recommended in the temporary use authorization protocol issued by the French national drug and medication agency (ANSM) in august. Nevertheless, further studies on larger retrospective data are needed to confirm our results and to determine the best therapeutic options in undergoing dialysis diagnosed with COVID-19.

Our report has several limitations. The small size of the samples weakens the strength of the comparison between COVID_Neg and COVID_Pos patients. However, serial samples were available for most of the patients. Anti-N but not anti-S IgG, were screened before vaccination, which may have biased the number of patients with history of SARS-CoV-2 infection. Indeed, anti-N IgGs wane faster than anti-S.[22]. Serological assay using N as target are thus slightly less sensitive than those using S.[22]Our study would also have been enriched by studying cellular immunity. The lack of viral genome sequencing may mislead our interpretation of the outbreak, as previously observed.[24] However, our conclusions would

remain the same even if some patients had acquired SARS-CoV-2 concomitantly outside of the unit outbreak.

Finally, our findings may not be generalizable to more recent variants such as Omicron. Indeed, emergence of variants harboring mutations within epitopes targeted by neutralizing antibodies

is associated with increase in breakthrough infection. On one hand it confirms the link between neutralization and vaccine efficacy, on the other hand, it stresses the need to re-evaluate of correlate of protection as new variants emerged. Importantly, the concept of an humoral correlate of protection will likely remains valid regardless of variant, but it is also likely that the levels of antibodies required for protection will rise as new variants emerged. Thus, an update of vaccine to elicit broadly neutralizing antibodies may be needed. In addition, the most recent variant Omicron escapes most of therapeutic monoclonal antibodies, including Casirivimab and Imdevimab used in our study.[25] Thus, the strategy presented here is predicted to be ineffective against Omicron. However, some mAbs, such as Sotrovimab, remain effective against Omicron, and other are under development. Thus, mAbs therapy now needs to be determined with regards of viral variant and available mAbs to remain effective.

In conclusion, our study confirms the correlation between anti-spike IgG levels and neutralization titers and shows that the level of anti-spike IgG titer measured one month after vaccination correlate to protection in hemodialysis patients. In this high-risk population, BNT162b2 vaccination did not prevent the outbreak but, alone or in combination with mAbs, prevented the development of severe forms of COVID-19.

CONFLICT OF INTEREST STATEMENT

The authors have nothing to declare

AUTHORS' CONTRIBUTIONS

IB, AS, FL and TB designed the study. IB, AS and TB wrote the manuscript. IB and AS collected the data. HS performed statistical analysis. ML performed serological analysis. DP, TS and OS performed viro-neutralization analysis. AuS, DP, AH, MD, BG, EF, BK, DJ, PP, CR, CA, ML and FL critically reviewed the manuscript. Each author contributed important intellectual content during manuscript drafting; all authors read and approved the final version of the manuscript.

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Table 1. Characteristics of hemodialysis patients infected with SARS-CoV-2 alpha variant during the outbreak

	Age	Sex	Nephropathy	Immuno-compromised	Anti-spike Ab (BAU/ml)*	Antibody neutralization titer*	Lymphocyte count *	BNT162b2 last injection-Positive RT-PCR (days)	Ct **	Hospitalized	mABs	Severity
Patient 1	48	F	Diabetic	No	3	7.5	2200	5	36	Yes	Yes	Mild
Patient 2	70	F	Unkown	Prednisone (Previous KTr, returned on dialysis in 04/2021)	2	7.5	1700	66	18	Yes	Yes	Mild
Patient 3	52	M	Diabetic	No	0	7.5	1000	Refused vaccination	18	Yes	Yes	Moderate
Patient 4	89	F	Myeloma cast nephropathy	Chemotherapy	4	19.9	700	68	35	Yes	Yes	Moderate
Patient 5	84	F	Diabetic	No	247	1770	600	68	20	No	No	Asymptomatic
Patient 6	58	F	Malformative uropathy	No	197	173.9	1000	68	35	No	No	Asymptomatic
Patient 7	85	M	AL Amyloidosis	Chemotherapy	18	75.4	600	68	34	Yes	No	Mild
Patient 8	50	F	Nephrosclerosis	No	0	7.5	1000	Refused vaccination	35	No	No	Asymptomatic
Patient 9	83	M	Nephrosclerosis	No	0	N/A	480	68	13	Yes	Yes	Mild
Patient 10	60	F	aHUS	Prednisone (Previous KTr, returned on dialysis in 04/2020)	18	127.8	1000	70	19	No	Yes	Mild
Patient 11	78	M	Nephrosclerosis	No	142	175.2	1200	75	16	No	Yes	Asymptomatic
Patient 12	64	M	Nephrosclerosis	Prednisone (Previous KTr, returned on dialysis in 10/2019)	170	371.4	2000	75	18	No	Yes	Asymptomatic
Patient 13	33	F	Lupus	Rituximab, MMF, Prednisone	30	88.6	400	55	18	No	Yes	Mild
Patient 14	76	M	Myeloma cast nephropathy	Chemotherapy	21	N/A	1300	75	17	No	Yes	Asymptomatic

*One month before the cluster; ** at diagnosis. Abbreviations: F: female; M: male; aHUS: atypical hemolytic uremic syndrome; KTr: kidney transplantation; mAb: monoclonal antibodies (a cocktail of two mAbs: Casirivimab and Imdevimab, 120 mg/mL (Roche)) was used. RT-PCR: real time polymerase chain reaction; Ct: cycle threshold. Pauci-symptomatic infections are defined by symptoms related to COVID-19 without oxygen therapy (fever, asthenia, anosmia). Mild and moderate forms of COVID-19 were defined according to the need of oxygen therapy: no oxygen and <4 L per min, respectively

Table 2. Clinical and biological characteristics of the cohort

	Total	COVID-19_Neg	COVID-19_Pos	*p*
N	53	39	14	-
Female, n (%)	23 (43)	15 (39)	8 (57)	0.23
Age, median [IQR]	64 [53-81]	64 [54-81]	67 [52-83]	0.8
Fully vaccinated with BNT16 2b2mRNA vaccine*	50 (94)	38 (97)	12 (86)	0.13
COVID-19 infection before vaccination, n(%)	11 (21)	9 (23)	2 (14)	0.5
Cause of nephropathy, n (%)				0.19
- Diabetic or vascular nephropathy	25 (47)	18 (46)	7 (50)	
- Glomerulopathy	5 (9)	3 (8)	2 (14)	
- MCN/amyloidosis	6 (11)	3 (8)	3 (21)	
- ADPKD	9 (17)	9 (23)	0 (0)	
- Others	8 (15)	6 (15)	2 (14)	
Comorbidities, n (%) :				
- Hypertension	50 (94)	36 (92)	14 (100)	0.29
- Diabetes Mellitus	20 (38)	14 (36)	6 (43)	0.65
- Obesity	3 (6)	1 (3)	2 (14)	-
- Smokers	18 (34)	12 (31)	6 (43)	0.5
Immunosuppressive treatment <6 months	15 (28)	8 (21)	7 (50)	0.046
- Previous kidney transplantation	8 (15)	5 (13)	3 (21)	0.42
- Hemopathy	6 (11)	3 (8)	3 (21)	0.07
- Auto-immune disease	1 (2)	0 (0)	1 (7)	-
Hemodialysis schedules				0.002
- MWF	27 (51)	15 (39)	12 (86)	
- TTS	26 (49)	24 (62)	2 (19)	
BMI, kg/m^2**, median [IQR]	21 [19-24]	21 [19-23]	21 [17-26]	0.9
Albuminemia, g/L, median [IQR]**	33 [29-37]	34 [31-38]	32 [28-35]	0.23
Lymphocyte count, Giga/L, median [IQR]**	1100 [900-1600]	1100 [900-1650]	1000[625-1275]	0.2
Gamma globulins, g/L, median [IQR]**	9 [6.4-11]	9 [6.4-11]	9 [6.4-10]	0.6
Positive hepatitis B serology***, n(%)	23 (43)	16 (41)	7 (50)	0.8

*One patient in the COVID-19_Neg group and two patients in the COVID-19_Pos group refused to be vaccinated, seven patients in the COVID-19_Neg group and two patients in the COVID-19_Pos group received one injection of BNT162b2 vaccine because of previous SARS-Cov-2 infection. **One month before COVID-19 outbreak. *** Hepatitis B serology was considered positive if IgG anti-HBs were superior to 10UI/L with or without anti-HBc. All patients were supposed to be vaccinated in the past. Abbreviations: MCN: myeloma cast nephropathy; ADPKD: autosomic dominant polycystic kidney disease; MWF: Monday-Wednesday-Friday dialysis schedule; TTS: Tuesday-Thursday-Saturday dialysis schedule; BMI:

body-mass index. Quantitative data are presented as median [interquartile range] and qualitative data as n(%), as appropriate.

Table 3. Clinical and biological characteristics of dialysis patients on MWF and TTS schedules

	Total	MWF	TTS	p
N	53	26	27	-
Fully vaccinated with BNT16 2b2mRNA vaccine*	50 (94)	24 (96)	25 (93)	>0.9
COVID-19 infection before vaccination	11 (21)	5 (19)	6 (22)	>0.9
Female, n (%)	23 (43)	12 (46)	11 (41)	>0.9
Age, median [IQR]	64 [53-81]	75 [52-82]	61 [54-78]	0.4
Cause of nephropathy - Diabetic or vascular nephropathy - Glomerulopathy - MCN/amyloidosis - ADPKD - Others	 25 (47) 5 (9) 6 (11) 9 (17) 8 (15)	 11 (42) 3 (12) 6 (23) 3 (12) 4 (15)	 14 (52) 2 (7) 0 (0) 6 (22) 4 (15)	0.1
Comorbidities, n (%) : - Hypertension - Diabetes Mellitus - Smokers	 50 (94) 20 (38) 18 (34)	 26 (100) 8 (31) 9 (35)	 24 (89) 12 (44) 9 (33)	 0.9 0.3 0.9
Immunosuppressive treatment <6 months - Previous kidney transplantation - Hemopathy - Auto-immune disease	15 (28) 8 (15) 6 (11) 1 (2)	12 (46) 5 (19) 6 (23) 1 (4)	3 (11) 3 (11) 0 (0) 0 (0)	0.01
BMI, kg/m^2**, median [IQR]	21 [19-24]	21 [16-24]	23 [20-25]	0.2
Albuminemia, g/L, median [IQR]**	33 [29-37]	32 [29-36]	35 [29-38]	0.3
Lymphocyte count, G/L, median [IQR]*	1100 [900-1600]	900 [700-1700]	1150 [1000-1625]	0.1
Positive hepatitis B serology, n(%)	23 (43)	12 (46)	10 (37)	0.8

*One month before COVID-19 outbreak. *Abbreviations: MCN: myeloma cast nephropathy; ADPKD: autosomic dominant polycystic kidney disease; MWF: Monday-Wednesday-Friday dialysis schedule; TTS: Tuesday-Thursday-Saturday dialysis schedule; BMI: body-mass index.* Quantitative data are presented as median [interquartile range] and qualitative data as n(%), as appropriate.

Table 4. Independent risk factors of SARS-CoV-2 infection during the outbreak

Characteristic	OR	95%CI	p
Anti-spike antibodies*			
<205 BAU/mL	Ref	-	-
>205 BAU/mL	0.16	[0.03-0.74]	0.025
Dialysis schedule			
TTS	Ref	-	-
MWF	7.37	[1.66-41.5]	0.013

205 BAU/mL represent the median of anti-S antibodies in the whole cohort, one month before the outbreak including the three unvaccinated patients; 7.1 BAU/mL (Binding Antibody Units) is equivalent to 50 AU/mL (Arbitrary units). Abbreviations: OR: odds ratio; CI: confident interval; TTS: Tuesday-Thursday-Saturday. All variables with a P value <0.2 in univariable analysis were included in the multivariable analysis. Stepwise backward selection based on the Akaike information criterion was used for the final multivariable model (logistic regression).
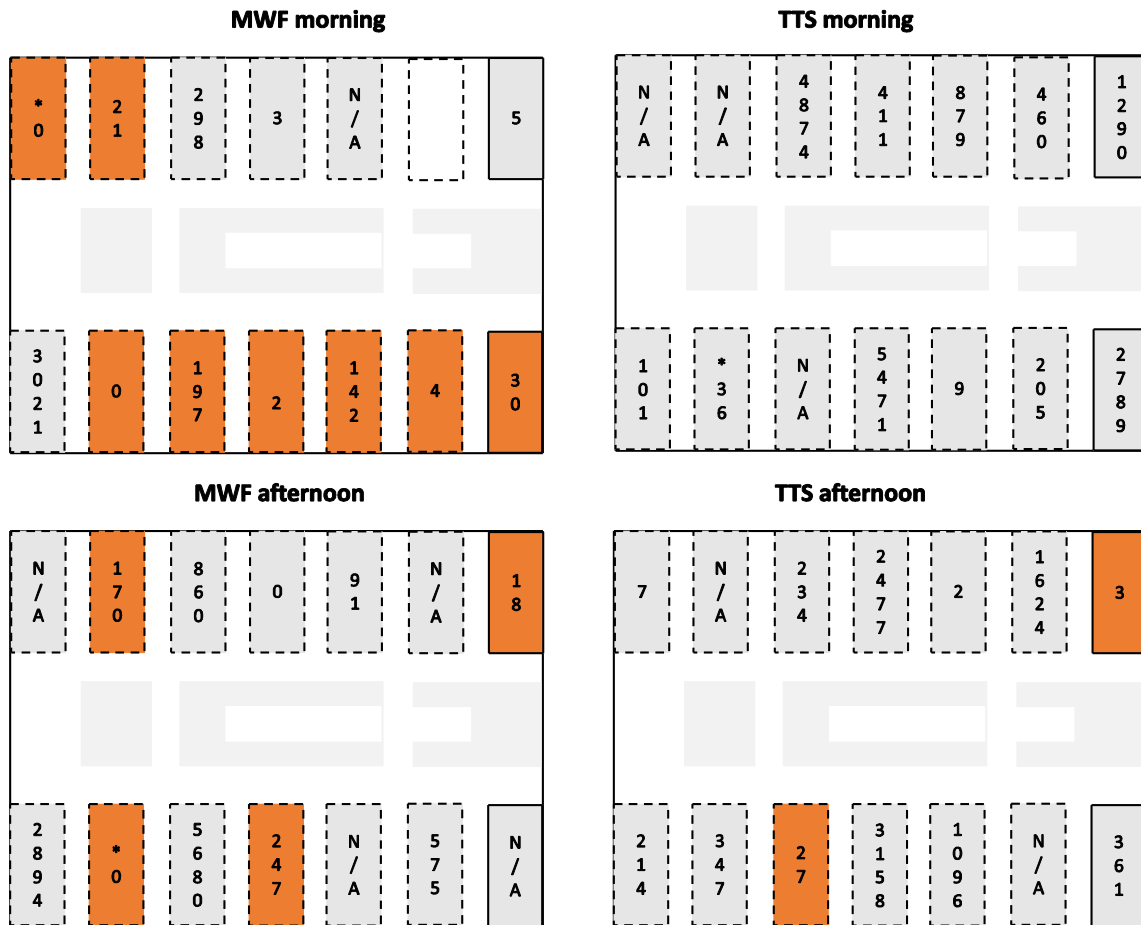
Figure 1. Representation of the dialysis room layout on Monday-Wednesday-Friday (left) and Tuesday-Thursday-Saturday (right) morning (up) and afternoon (down) schedules, when the COVID-19 outbreak was diagnosed. Anti-spike antibody titers, one month before the cluster are shown in each rectangle (BAU/L). *7.1 BAU/mL (*Binding Antibody Units]) is equivalent *to 50 AU/mL (*Arbitrary units]). Gray rectangles represent the patients who were not infected with SARS-CoV-2 during the outbreak. Orange rectangles represent the patients infected with SARS-CoV-2 during the outbreak. *non-vaccinated; Abbreviations: MWF: Monday-Wesdnesday-Friday; TTS: Tuesday-Thursday-Saturday. N/A: serology non available. Note that the rooms at the left and right corners are closed while the other beds are in an open room.
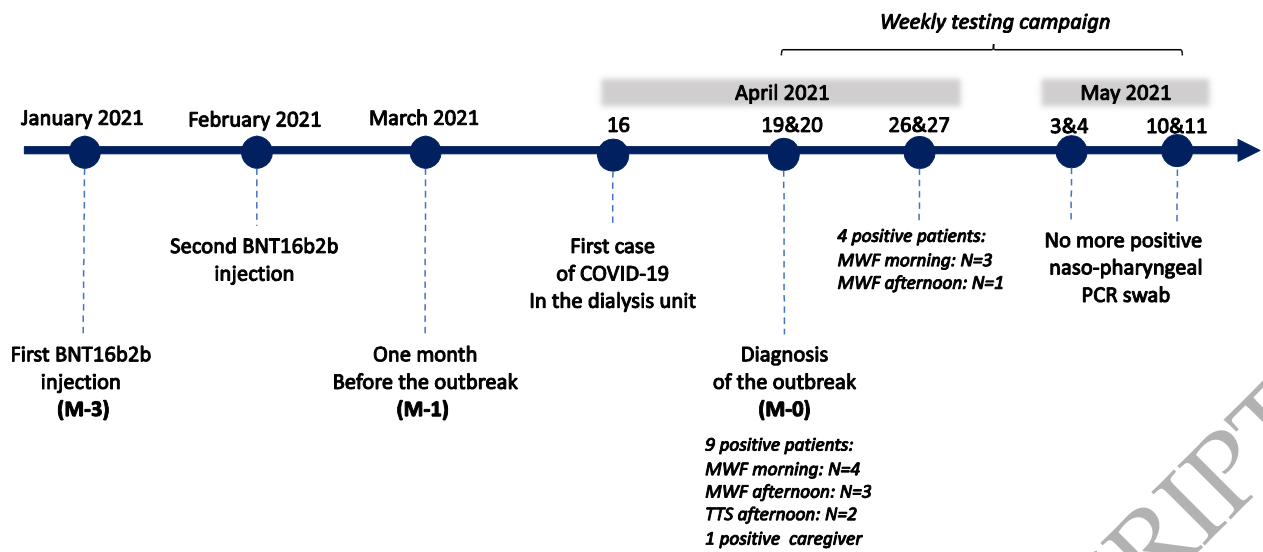
Figure 2: Timeline of the vaccination campaign followed by SARS-CoV-2 alpha variant outbreak in the dialysis unit. Vaccination campaign began in January and ended in March. Serologies were analyzed before vaccination (M-3 timepoint), one month before the outbreak (M-1 time point) and at the beginning of the outbreak (M-0 timepoint) *Abbreviations: MWF: Monday, Wednesday, Friday shift; TTS: Tuesday, Thursday, Saturday shift. PCR: polymerase chain reaction*
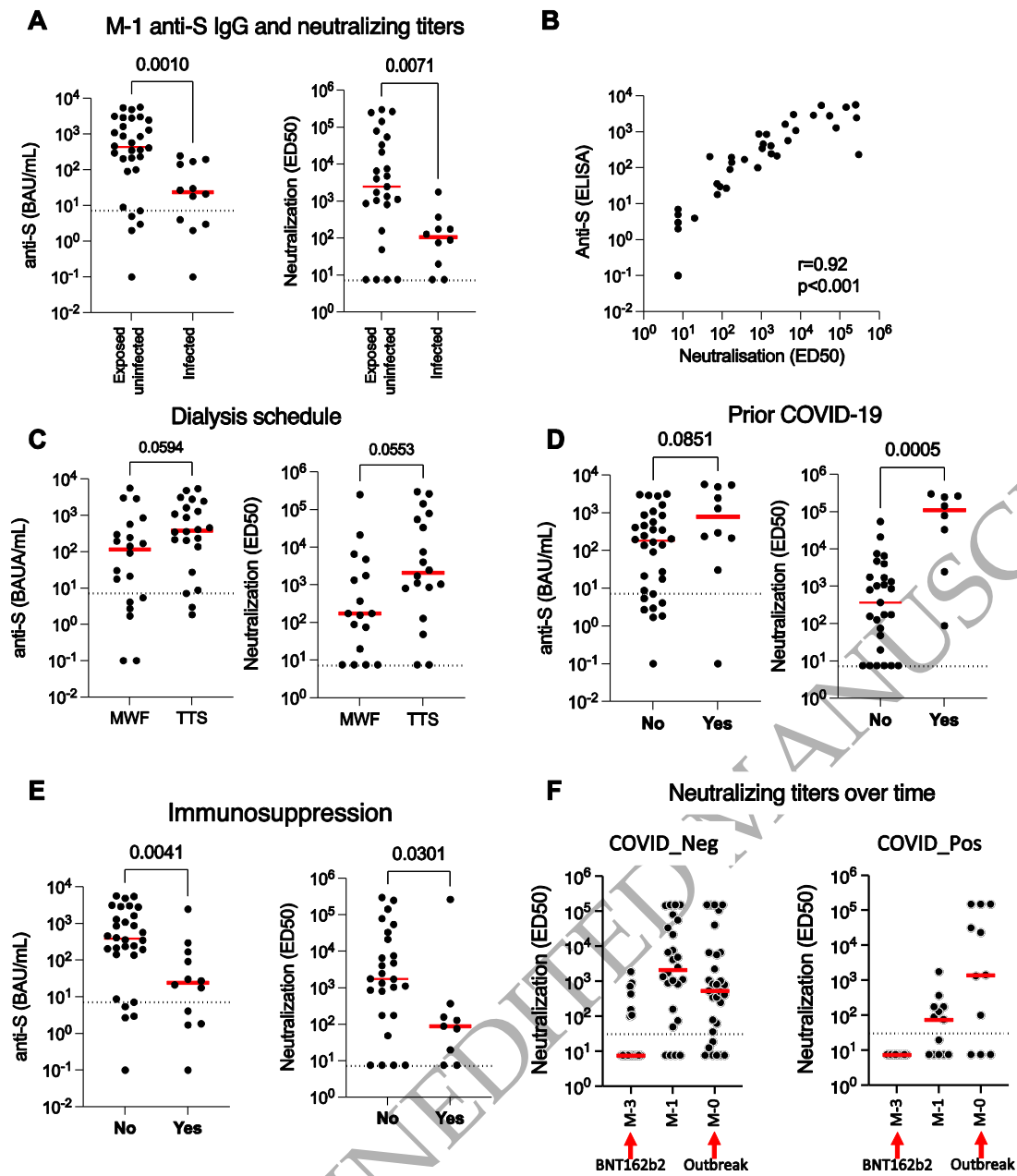
Figure 3: Characterization of the humoral immunity of dialyzed patients prior to the outbreak. A) Anti-S antibodies (left, BAU/mL) and neutralizing (right) titers in sera of individuals exposed to (COVID-19 -) or infected with (COVID-19 +) SARS-CoV-2. p- value is determined using a Mann-Whitney test. **B)** Correlation between neutralizing titers and anti-S antibody quantifications in sera of all individuals. r coefficient and p- value are determined

using a Spearman correlation test. **C)** Anti-S antibodies (left, BAU/mL) and neutralizing (right) titers in sera of individuals dialyzed on MWF or TTS schedule. p- value is determined using a Mann-Whitney test. **D)** Anti-S antibodies (left, BAU/mL) and neutralizing (right) titers in sera of individuals with (Yes) or withour (No) prior infection with SARS-Cov-2 before the outbreak. p- value is determined using a Mann-Whitney test. **E)** Anti-S antibodies (left, BAU/mL) and neutralizing (right) titers in sera of individuals with (Yes) or without (No) an additional cause of immunosuppression. p- value is determined using a Mann-Whitney test. **F)** Neutralizing titers in sera of individuals exposed to (COVID-19_Neg, left) or infected with (COVID-19_Pos, right) SARS-CoV-2, before vaccination (M-3), one month before the outbreak (M-1) and at the beginning of the outbreak (M-0). *MWF: Monday-Tuesday-Friday; TTS: Tuesday-Thursday-Wednesday*.