



ORIGINAL ARTICLE

Comparison of antibody response to SARS-CoV-2 after two doses of inactivated virus and BNT162b2 mRNA vaccines in kidney transplant

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ABSTRACT

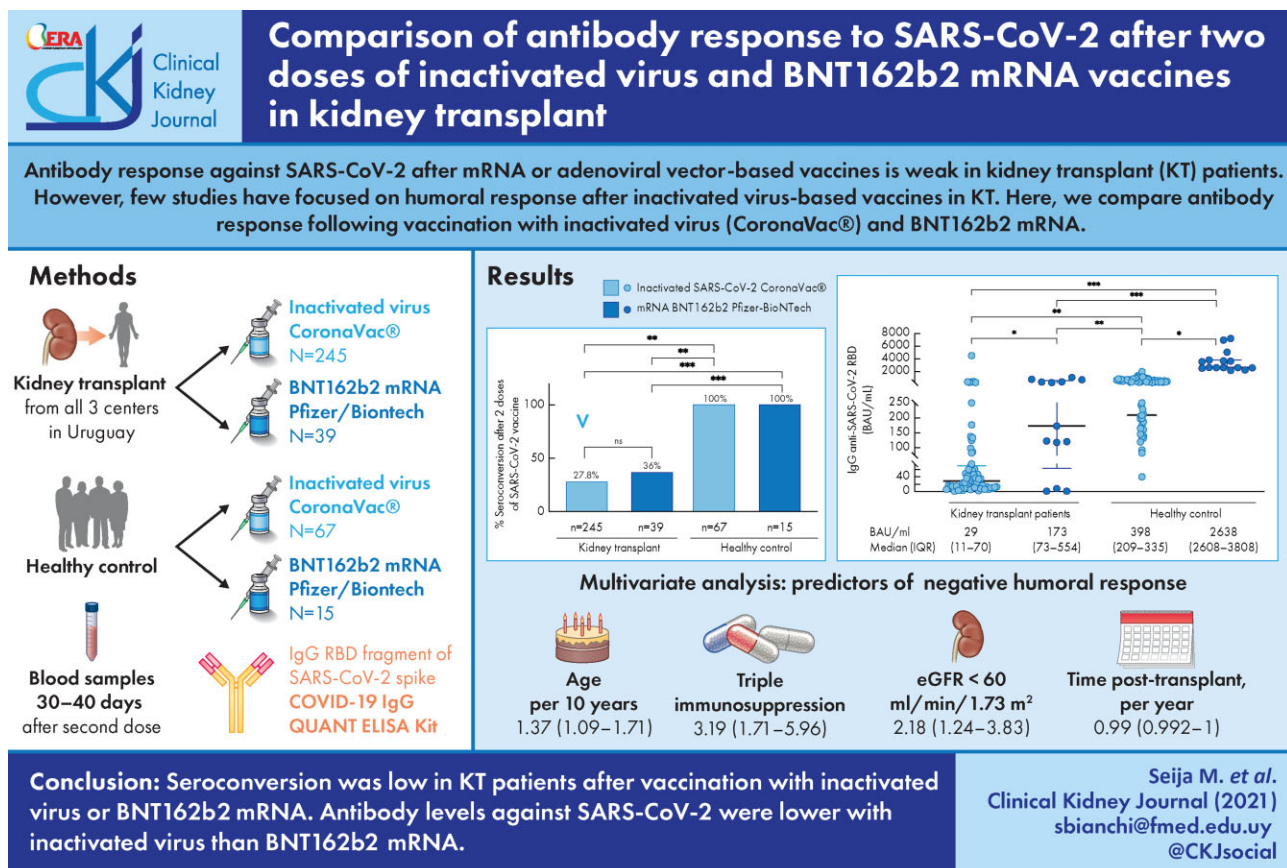
Background. Antibody response against severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) after mRNA or adenoviral vector-based vaccines is weak in kidney transplant (KT) patients. However, few studies have focused on humoral response after inactivated virus-based vaccines in KT. Here, we compare antibody response following vaccination with inactivated virus (CoronaVac®) and BNT162b2 mRNA.

Methods. A national multicentre cross-sectional study was conducted. The study group was composed of patients from all KT centres in Uruguay, vaccinated between 1 and 31 May 2021 (CoronaVac®, $n = 245$ and BNT162b2, $n = 39$). The control group was constituted of 82 healthy individuals. Participants had no prior confirmed coronavirus disease 2019 (COVID-19) test. Blood samples were collected between 30 and 40 days after the second dose. Serum-specific immunoglobulin G (IgG) antibodies against the receptor-binding domain (RBD) of SARS-CoV-2 Spike protein were determined using the COVID-19 IgG QUANT ELISA Kit.

Results. Only 29% of KT recipients showed seroconversion (36.5% BNT162b2, 27.8% inactivated virus, $P = 0.248$) in comparison with 100% in healthy control with either vaccine. Antibody levels against RBD were higher with BNT162b2 mRNA than with inactivated virus [median (interquartile range) 173 (73–554) and 29 (11–70) binding antibody units (BAU)/mL, $P < 0.034$] in KT and 10 times lower than healthy control [inactivated virus: 308 (209–335) and BNT162b2: 2638 (2608–3808) BAU/mL, $P < 0.034$]. In multivariate analysis, variables associated with negative humoral response were age, triple immunosuppression, estimated glomerular filtration rate and time post-KT.

Conclusion. Seroconversion was low in KT patients after vaccination with both platforms. Antibody levels against SARS-CoV-2 were lower with inactivated virus than BNT162b2 mRNA. These findings support the need for strategies to improve immunogenicity in KT recipients after two doses of either vaccine.

GRAPHICAL ABSTRACT



Keywords: COVID-19, kidney transplantation, SARS-CoV-2 vaccine

INTRODUCTION

Kidney transplant (KT) patients are at high risk of severe severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection [1–5]. Hence, vaccination in this population is recommended. Recent evidence suggests that antibody response among solid organ transplant (SOT) patients is weak after two doses of mRNA-based vaccine or adenoviral vector platform [6–23]. However, only one study has addressed humoral response after inactivated virus-based vaccine in KT patients [24].

Inactivated virus vaccines are a well-known technology and have several advantages for large-scale utilization, including their stability at non-extreme refrigeration temperatures and their long lifespan [25]. These characteristics make them a useful tool in the global fight against coronavirus disease 2019 (COVID-19), although more data are needed on its efficacy in KT recipients. Furthermore, recently it has been approved by the European Medicine Agency for emergency use in Europe and to date, more than 750 000 000 have been administered in more than 40 countries [26, 27].

In Uruguay, the Ministry of Public Health approved inactivated SARS-CoV-2 (CoronaVac®, Sinovac Biotech Ltd) and BNT162b2 mRNA (Pfizer/BioNTech) vaccines for emergency use [28, 29]. Healthcare workers were vaccinated with BNT162b2 mRNA. SOT patients were prioritized and vaccinated according to age group (people between 18 and 70 years received inactivated virus-based and those over 70 years were vaccinated with BNT162b2 mRNA). CoronaVac® was administered to almost 70% of the Uruguayan population, including SOT patients, with a reduction of infection and intensive care admissions according to the Health Ministry Authority.

The aim of this work was to compare humoral response after inactivated virus and BNT162b2 mRNA vaccines in KT patients and evaluate adverse events associated with vaccination.

MATERIALS AND METHODS

We conducted a national multicentre cross-sectional study to evaluate humoral response after SARS-CoV-2 vaccination in KT patients. The study group was composed of KT recipients from all KT centres in Uruguay (INU-Hospital Italiano, Hospital Evangélico and Hospital de Clínicas). Inclusion criteria were age >18 years old, kidney or kidney-pancreas transplant, no prior confirmed COVID-19 and had a second dose of either vaccine, BNT162b2 mRNA or inactivated virus (CoronaVac®), between 1 and 31 May. The control group was constituted by 82 healthy individuals [40.2% men; median age 41 years, interquartile range (IQR) 33–49], without immunosuppression medication, no prior confirmed COVID-19 and have the second dose of either vaccine BNT162b2 mRNA (15 individuals, 40% men; median age 42 years, IQR 37–47) or inactivated virus (CoronaVac®) (67 individuals, 40.3% men; median age 41 years, IQR 30–49).

All subjects received two doses (30 µg each) of BNT162b2 mRNA or two doses (600 SU each) of inactivated virus, both 28 days apart, according to the recommendations of the Uruguayan National Health Authority. None of the participants had prior confirmed COVID-19. Clinical data were recorded. A questionnaire of adverse events was performed, including all symptoms 7 days after each dose, which included local side effects such as pain, redness, swelling and regional lymphadenopathy, and systemic side effects such as fever, chills, headache, fatigue, myalgia, arthralgia, nausea, vomiting and diarrhoea. Estimated glomerular filtration rate (eGFR) was calcu-

lated using the Chronic Kidney Disease Epidemiology Collaboration (CKD-EPI) formula.

Blood samples were collected between 30 and 40 days after the second dose injection. Freshly collected blood in clot activator and gel tube was centrifuged (2500 r.p.m., 15 min). Sera were separated and stored at –20°C until analysis.

The level of serum-specific Immunoglobulin G (IgG) antibodies against the receptor-binding domain (RBD) of SARS-CoV-2 Spike protein was determined using the COVID-19 IgG QUANT ELISA Kit (developed by Universidad de la República, Institut Pasteur de Montevideo and ATGen Company), according to manufacturer's instruction. The assay has a sensitivity of 97.7% and specificity 96.2%. Quantitative test results were expressed in binding antibody units (BAU)/mL referring to the First WHO International Standard for anti-SARS-CoV-2 immunoglobulin (NIBSC code: 20/136) used for assay calibration. Seroconversion was defined as presence of specific IgG antibodies against the RBD.

The study was approved by the ethical institutional review board (MSP 3535533-956220).

Continuous variables were tested for normal distribution (Kolmogorov-Smirnov). Normally distributed variables were expressed as mean ± standard deviation (SD), non-normally distributed as median and interquartile range, and qualitative variables as number and percentage. Categorical variables were compared using Chi-squared statistic. Continuous variables were compared using t-test (normally distributed) or Kruskal-Wallis/Mann-Whitney (non-normally distributed).

Group analysis included two groups according to SARS-CoV-2 IgG antibodies status: positive or negative.

Binary logistic regression models for negative serology test risk were fitted including the significant variables in univariate analysis. $P < 0.05$ was considered statistically significant. IBM® SPSS® version 22 (Chicago, IL, USA) statistical software was used for statistical analyses and Graph Pad 8 for charts constructs.

RESULTS

Among 1400 KT patients in Uruguay, 284 KT recipients were included in this study. In addition, 82 healthy individuals were included as a control group. Both groups underwent serological testing for SARS-CoV-2-specific antibodies after two doses of inactivated virus or BNT162b2 mRNA vaccine. Baseline patients' characteristics are detailed in Table 1. Seroconversion was lower in KT patients than healthy control (29% versus 100%). Seroconversion in KT with BNT162b2 mRNA (36.5%) was higher than inactivated virus (27.8%), but without statistical significance ($P = 0.248$, Figure 1A).

Serum levels of anti-RBD IgGs were significantly higher in KT patients who received BNT162b2 mRNA compared with inactivated virus vaccine, with a median of 173 (73–554) and 29 (11–70) BAU/mL respectively ($P < 0.034$, Figure 1B). Compared with the healthy control group, KT had lower levels of antibody with either vaccine.

Seropositive patients for anti-RBD were significantly younger, had higher eGFR and lymphocyte count, and longer time since transplantation. With regard to immunosuppression treatment, these patients were less frequently on triple therapy (antimetabolite, calcineurin inhibitor and prednisone) and more often on everolimus treatment (Table 1).

In multivariate analysis, variables associated with negative humoral response were age [per 10 years, odds ratio 1.372 [95% confidence interval (CI) 1.097–1.715], $P = 0.006$], triple immunosuppression [3.197 (1.714–5.96), $P = 0.000$] and eGFR

Table 1. Clinical characteristics of patients according to IgG anti-RBD SARS-CoV-2 status after inactivated SARS-CoV-2 or BNT162b2 mRNA vaccine

Variable	IgG anti-RBD SARS-CoV-2		Total	P-value
	Negative	Positive		
N, (%)	204 (71)	80 (29)	284 (100)	
Type of vaccine, n (%)				
Inactivated SARS-CoV-2	179 (87.7)	66 (82.5)	245 (86.3)	0.335
mRNA BNT162b2	25 (12.5)	14 (17.5)	39 (17.3)	
Age years, median (IQR)	56 (45–73)	52 (39–72)	55 (43–72)	0.039
Sex, men n (%)	119 (59.5)	46 (58)	165 (59)	0.759
Comorbidities, n (%)				
Stroke	7 (4.5)	1 (1.8)	8 (3.8)	0.369
Ischaemic heart disease	17 (11)	4 (7.3)	21 (10)	0.433
Peripheral arteriopathy	4 (2.6)	0 (0)	4 (1.9)	0.229
Diabetes mellitus, n (%)	61 (30.2)	16 (20.3)	77 (27.4)	0.093
BMI	26.3 (23.1–35)	25.3 (22.3–35.2)	26 (22–35)	0.187
Type of transplant, n (%)				
Kidney	194 (95)	78 (98)	272 (95)	0.365
Kidney–pancreas	10 (5)	2 (2)	12 (4.2)	
Time of transplant months, median (IQR)	57 (29–221)	76 (37–263)	61 (32–230)	0.061
Patients in the first year of transplant, n (%)	19 (9.4)	7 (8.9)	26 (9.3)	0.887
Triple immunosuppression, n (%)	167 (82.7)	48 (68.8)	215 (76.5)	0.000
Antimetabolite, n (%)				0.000
None	19 (9.5)	21 (25.6)	40 (14.3)	0.023
Mycophenolate	178 (88.6)	48 (60.8)	226 (80.7)	
Azathioprine	4 (2)	10 (12.7)	14 (5)	
Calcineurin inhibitors, n (%)				
None	5 (2.5)	8 (10.1)	13 (4.6)	0.111
Tacrolimus	166 (82.2)	60 (75.9)	226 (80.4)	
Cyclosporine	31 (15.3)	11 (13.9)	42 (16.9)	
Prednisone, n (%)	192 (95)	71 (89.9)	263 (93.6)	0.000
Everolimus, n (%)	18 (9)	22 (27.8)	40 (14.3)	0.274
Rituximab, n (%)	3 (1.5)	0 (0)	3 (1.1)	0.477
Thymoglobulin, n (%)	29 (14.6)	9 (11.4)	38 (13.7)	0.838
Rejection in last 3 months, n (%)	6 (3)	2 (2)	8 (2.9)	0.09
Lymphocyte count, cells/ μ L, median (IQR)	1900 (1400–3696)	2213 (1740–4200)	2000 (1454–3820)	0.020
Serum creatinine μ mol/L, median (IQR)	124 (101–1414)	112 (91–241)	120 (97–352)	0.011
eGFR mL/min/1.73 m ² , mean \pm SD	50.3 \pm 23	58.4 \pm 22	52 \pm 22	

BMI, body mass index; triple immunosuppression, antimetabolite + calcineurin inhibitor + prednisone.

Table 2. Binary logistic regression model of predictors of negative humoral response 30 days after two doses of inactivated SARS-CoV-2 or BNT162b2 mRNA vaccine

Predictor	Odd ratio	95% CI	P-value
Immunosuppression			
Other	Ref		
Triple immunosuppression	3.197	1.714–5.962	0.000
Age, per 10 years old	1.372	1.097–1.715	0.006
eGFR			
≥ 60 mL/min/1.73 m ²	Ref		
< 60 mL/min/1.73 m ²	2.184	1.243–3.838	0.007
Time after KT, per year	0.996	0.992–1.000	0.034

Triple immunosuppression, antimetabolite + calcineurin inhibitor + prednisone.

< 60 mL/min/1.73 m² [2.18 (1.24–23.83), $P = 0.007$], whereas time post-KT was negatively associated with negative humoral response [0.996 (0.992–1)] (Table 2).

The most frequent adverse event with each vaccine dose was pain at the site of injection, with significant difference between platforms (24% CoronaVac[®] and 48% mRNA, $P < 0.05$). Other less

frequent events were headache, arthralgia, myalgia and pruritus, without difference in frequency comparing the first with the second dose. There were no severe side-effects reported in this cohort (Figure 2).

DISCUSSION

Prior studies have shown low seroconversion after two doses of mRNA and viral vector-based vaccines [6–23]. To our knowledge, this is the first study that analyses the humoral response after two doses of inactivated SARS-CoV-2 vaccine in KT patients. Only 29% of KT recipients had antibody response against SARS-CoV-2 vaccine, in contrast to 100% of healthy controls. There was no difference in seroconversion between the two vaccine platforms analysed. Nonetheless, mean antibody titres were higher with mRNA-based vaccine than with inactivated SARS-CoV-2 platform.

These findings agree with previous studies that report a seroconversion between 10% and 40% with mRNA-based vaccine [6–8, 17–23]. In concordance with other studies, mean antibody titres in KT patients were up to 10 times lower than healthy controls [17]. There is only one study that

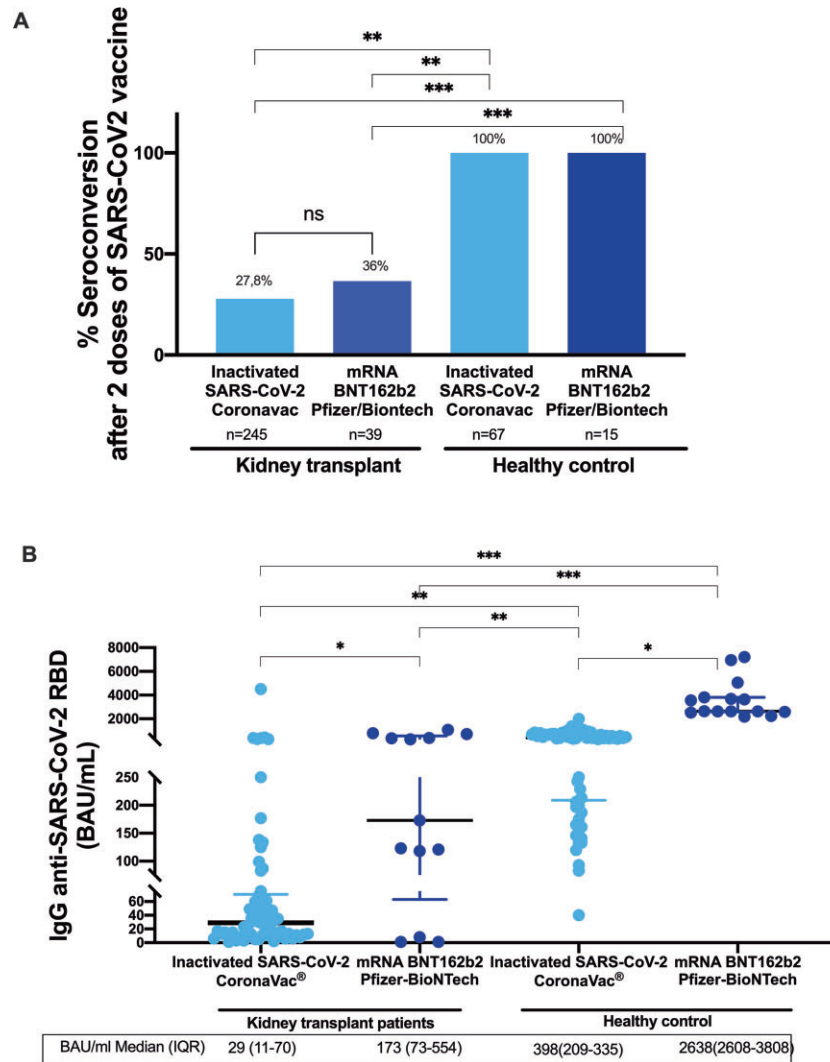


FIGURE 1: Serological response after two doses of inactivated SARS-CoV-2 or BNT162b2 mRNA vaccine in kidney transplant patients and healthy control. (A) Percentage of seroconversion. (B) IgG anti-RBD SARS-CoV-2 titres (BAU/mL) in patients with seroconversion. * $P < 0.05$ mRNA versus inactivated SARS-CoV-2, ** $P < 0.05$ versus inactivated SARS-CoV-2 in healthy control, *** $P < 0.05$ versus BNT162b2 mRNA in healthy control, ns, not significant.

compares antibody titres between two different vaccine platforms in KT patients, showing enhanced humoral responses with BNT162b2 mRNA-based compared with ChAdOx1-based vaccine. The clinical significance of this finding should be further evaluated [8].

The most important risk factors for no serological response to vaccines are associated with net immunosuppression such as age, triple immunosuppression, tacrolimus and mycophenolate, low lymphocyte count and eGFR.

Our results provide additional evidence of a weak immune response with two different vaccine platforms. To reinforce immunity, higher and/or supplemental booster doses are the more common solutions. Recently, it has been reported that a third and a fourth dose of BNT162b2 mRNA to SOT recipients improved immunogenicity [30–32]. There is growing evidence that combination of different platforms enhanced immunity [33].

A potential bias for this study was the criteria to receive each platform vaccine in Uruguay. BNT162b2 mRNA-vaccinated

group had small size and included healthcare workers from all ages and patients older than 70 years. However, since most patients were vaccinated with inactivated virus vaccine, this work contributes to understanding the humoral response after inactivated SARS-CoV-2 vaccine in KT population. Another drawback of this work could be the absence of anti-RBD IgGs determination before vaccination in these patients.

In conclusion, we report a weak humoral response after two doses of inactivated SARS-CoV-2 or BNT162b2 mRNA vaccine in KT recipients (29% of seroconversion). IgG antibody titres in KT were 10 times lower than healthy controls, even though they were higher with mRNA vaccine. Further study is needed to determine the impact of COVID-19 in these patients. Different strategies could improve immunogenicity, such as additional doses or a combination of platforms [30–32, 34, 35]. Based on these findings, we strongly recommend that all transplant recipients should continue with the non-pharmacological protection measures, including masks, hand hygiene and social distancing.

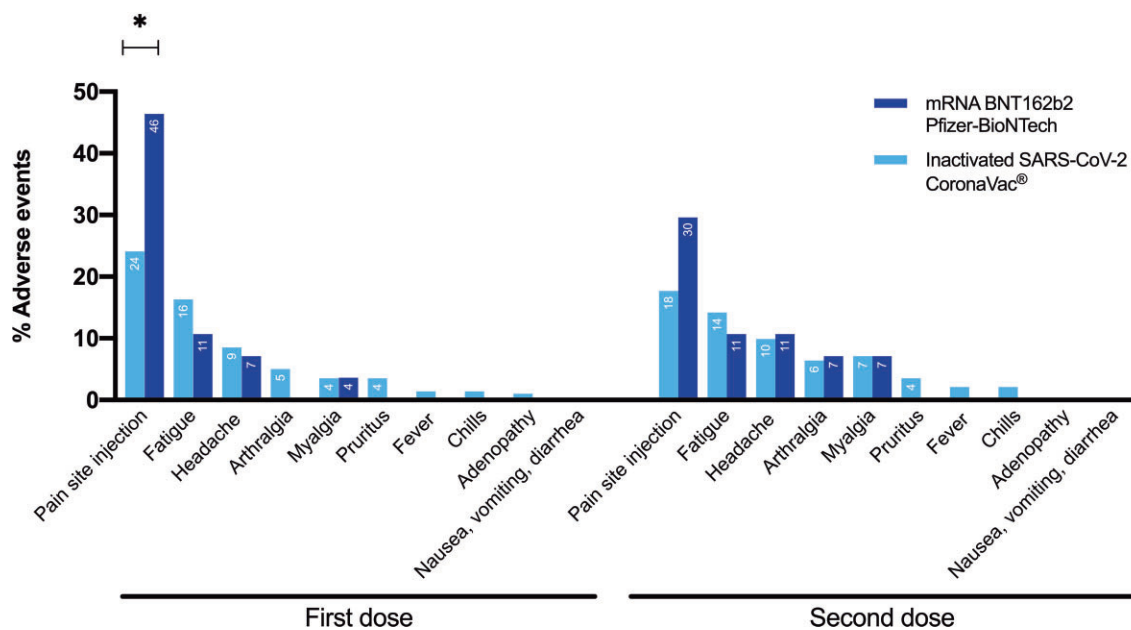


FIGURE 2: Side effects with first and second dose of inactivated SARS-CoV-2 or BNT162b2 mRNA vaccine. * $P < 0.05$ mRNA versus inactivated SARS-CoV-2.

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This protocol was approved by Hospital de Clínicas, Facultad de Medicina, Universidad de la República Ethics Committee.

CONFLICT OF INTEREST STATEMENT

The authors of this manuscript have no conflict of interest to disclose.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available on reasonable request from the corresponding author S.B., or M.S. The data are not publicly available due to restrictions for containing information that could compromise the privacy of research participants.

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