

Antibody response after one and two doses of the BNT162b2 vaccine in nursing home residents: The CONSORT-19 study

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Abstract

Methods: Twenty-two French nursing homes were included. COVID-19 had been diagnosed with real-time reverse-transcriptase polymerase chain reaction (RT-PCR) for SARS-CoV-2. Blood S-protein IgG and nucleocapsid (N) IgG protein (N-protein IgG) were measured 21-24 days after the first jab (1,004 residents) and 6 weeks after the second (820 residents). **Results:** Among the 735 residents without prior COVID-19, 41.7% remained seronegative for S-protein IgG after the first jab vs 2.1% of the 270 residents with a previous positive RT-PCR ($p < 0.001$). After the second jab, only 3% of the 586 residents without prior COVID-19 remained seronegative. However, 26.5% of them had low S-protein IgG levels (50-1050 UA/mL) vs 6.4% of the 222 residents with prior COVID-19. Residents with old infection (first wave), or seropositive for N-protein IgG at the time of vaccination, had the highest S-protein IgG levels. Residents with a prior COVID-19 infection had higher S-protein IgG levels after one dose than those without two jabs. **Interpretation:** A single vaccine jab is sufficient to reach immunity in residents with prior COVID-19. Most residents without prior COVID-19 are seropositive for S-protein IgG after the second jab, but around 30% have low levels of S-protein IgG. Whether residents with no or low post-vaccine immunity are at higher risk of symptomatic COVID-19 requires further analysis.

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Running title: Antibody level after BNT162b2 vaccine

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Summary of the article's main point:

One BNT162b2 jab is sufficient for immunity in 98% of nursing home residents with prior COVID-19 and in 53.8% of residents without. After two jabs, levels of S-protein IgG are undetectable or low in 3.1% and 26.3% of residents with and without prior COVID-19, respectively.

Abstract

Background: There is insufficient information on the humoral immune response following COVID-19 vaccination in nursing home residents. A longitudinal study compared SARS-CoV-2 spike IgG (S-protein IgG) levels after one and two BNT162b2/Pfizer jabs in residents with and without prior COVID-19.

Methods: Twenty-two French nursing homes were included. COVID-19 had been diagnosed with real-time reverse-transcriptase polymerase chain reaction (RT-PCR) for SARS-CoV-2. Blood S-protein IgG and nucleocapsid (N) IgG protein (N-protein IgG) were measured 21-24 days after the first jab (1,004 residents) and 6 weeks after the second (820 residents).

Results: Among the 735 residents without prior COVID-19, 41.7% remained seronegative for S-protein IgG after the first jab vs 2.1% of the 270 residents with a previous positive RT-PCR ($p < 0.001$). After the second jab, only 3% of the 586 residents without prior COVID-19 remained seronegative. However, 26.5% of them had low S-protein IgG levels (50-1050 UA/mL) vs 6.4% of the 222 residents with prior COVID-19. Residents with old infection (first wave), or seropositive for N-protein IgG at the time of vaccination, had the highest S-protein IgG levels. Residents with a prior COVID-19 infection had higher S-protein IgG levels after one dose than those without two jabs.

Interpretation: A single vaccine jab is sufficient to reach immunity in residents with prior COVID-19. Most residents without prior COVID-19 are seropositive for S-protein IgG after the second jab, but around 30% have low levels of S-protein IgG. Whether residents with no or low post-vaccine immunity are at higher risk of symptomatic COVID-19 requires further analysis.

Key words: COVID-19, BNT162b2/Pfizer vaccine, nursing home, rRT-PCR, antibodies against SARS-CoV-2

Background

Nursing Home (NH) residents are at high risk of serious illness and death from COVID-19 due to Severe Acute Respiratory Syndrome Coronavirus-2 (SARS-CoV-2) ¹. In NHs facing a COVID-19 outbreak, infection prevention and control (IPC) measures based on results of repeated testing by real-time reverse-transcriptase polymerase chain reaction for SARS-CoV-2 (RT-PCR) in all residents can limit coronavirus transmission ^{2,3}. Residents with a prior COVID-19 may be protected against reinfection during outbreaks in NHs ⁴.

Vaccination against SARS-CoV-2 is safe and effective in preventing COVID-19 in adults ⁵, and immunocompetent SARS-CoV-2 seropositive adults might only require one dose of the Pfizer-BioNTech or the Moderna mRNA vaccines to reach immunity ^{6,7}.

Age or immune dysregulation may impact immune response to COVID-19 and vaccination. The level of N-protein IgG produced after SARS-CoV-2 infection, associated with a substantially reduced risk of SARS-CoV-2 reinfection ⁸, declines over time ⁹. In the Danish Microbiology Database, protection against repeat infection was found to be reduced in participants over 65 years ¹⁰. Age seems also to reduce serological response after vaccination of kidney ¹¹ or liver transplant recipients ¹², and of patients with chronic lymphocytic leukemia, ¹³ cancer ¹⁴ or hemodialysis. ¹⁵

Few data exist on post-vaccine immunity in NH residents, and the size of the study is usually low. After one jab, binding antibodies against S-protein were approximately 4-fold lower in residents compared to healthcare professionals. ^{16,17} Although the increment of antibody levels after the second immunisation may be higher

in older participants, the absolute mean titer of this group seems to remain lower in NH residents aged over 80 years than in those under 60.¹⁸

A pilot study showed that NH residents having had COVID-19 in the past 9-12 months had much higher levels of antibodies against the SARS-CoV-2 S-protein (S-protein IgG) after a single BNT162b2 jab than residents not having had COVID-19¹⁶.

It remains therefore necessary *i*) to confirm in a large sample of residents whether the S-protein IgG response after a single BNT162b2 jab may be influenced by prior COVID-19 infection, *ii*) to determine in former COVID-19 residents whether the S-protein IgG response after a single BNT162b2 jab may be influenced by the time elapsed since the COVID-19 infection and by the persistence of a natural immunity against N-protein and (iii) to assess the S-protein IgG response after a second vaccination in residents with or without prior COVID-19.

We first compared S- and N-protein IgG levels 3 weeks after a single BNT162b2 jab in a large group of residents with vs without prior COVID-19 confirmed by RT-PCR (RT-PCR-positive residents). Two periods were considered - the past 9 to 12 months (older infections) and the past 3 to 7 months (newer infections) - corresponding to the first and second waves of the epidemic in our region (March-June 2020 and September-December 2020). Secondly, we assessed the S-protein IgG response in these residents 6 weeks after a second vaccination.

Methods

Design

A longitudinal study was carried out in NHs having faced a COVID-19 outbreak in 2020. Firstly, we compared N- and S-protein IgG levels three weeks after a single BNT162b2 jab in residents without prior COVID-19 (with repeated negative RT-PCR and negative N-protein IgG measured 3 weeks after the jab) and in residents with prior COVID-19 (confirmed either by a positive RT-PCR or by detectable N-protein IgG). Two periods were considered - the past 9 to 12 months (older infections) and the past 3 to 7 months (newer infections) - corresponding to the first and second waves of the epidemic in our region.

Secondly, S-protein levels were compared six weeks after a second BNT162b2 jab in residents with or without prior COVID-19.

Settings

We included 22 NHs of the Montpellier area (France), each with at least five residents with a previous diagnosis of COVID-19 confirmed by RT-PCR on a nasopharyngeal swab test between March and December 2020 (PCR-positive residents). This study follows previous studies conducted in the same NHs with a COVID-19 outbreak between March and June 2020^{16,19,20}.

Participants

As previously reported^{16,19,20}, as soon as a resident developed COVID-19 in a NH, all residents and staff members were repeatedly tested using RT-PCR on nasopharyngeal swabs until no new cases were diagnosed. This was the procedure recommended by the Health Agency of our region, in accordance with the European Geriatric Medicine Society guidance³.

All residents who had not had COVID-19 in the last 3 months, and who signed an informed consent, were offered a first jab of the BNT162b2 vaccine between January and March 2021. According to the national recommendations in NHs, a second jab, irrespective of prior COVID-19 history, was offered three weeks later. Residents and their family, relatives or legal representative were informed of the possibility to measure the antibody response after the first and second jabs and of the fact that the residents' anonymised clinical and biological data would be used for research purposes. The study was approved by the Montpellier University Hospital institutional review board (IRB-MTP_2020_06_202000534 and IRB-MTP_2021_04_202000534).

Outcomes

S-protein IgG against the SARS-CoV-2 Receptor-Binding Domain (RBD) of the S1 subunit was detected using the SARS-CoV-2 IgG II Quant assay (Abbott Diagnostics). Results were expressed as arbitrary units per mL (AU/mL; positive threshold: 50 AU/mL; upper limit: 40,000 AU/mL; a level [?] 1,050 AU/mL was considered as a significant response²¹ and a level [?] 4160 AU/mL as a level indicating a high neutralising effect according to the manufacturer). N-protein IgG were detected using the SARS-CoV-2 IgG assay (Abbott Diagnostics). Results were expressed as signal to cutoff ratio (S/CO; Abbott Alinity; positive threshold: 0.8 S/CO)²².

Sample size

The sample size was not calculated since we aimed to study all possible NH residents.

Statistical analysis

Qualitative variables were described with frequency and proportions for each category. The description of quantitative variables was performed using mean and standard deviation and/or median, minimum and maximum values. N-protein and S-protein IgG levels were compared in residents *i*) with and without prior COVID-19, *ii*) with prior COVID-19 during the first or second wave of the epidemic, and *iii*) between those who remained or did not remain seropositive for N-protein IgG. Wilcoxon–Mann–Whitney 2-sided tests were used, qualitative variables were compared using a chi-2 test, and the statistical significance threshold was set at 5%. Analyses were performed using the SAS Enterprise Guide, v7.3 (SAS Institute Inc.).

Results

Demographic characteristics of the residents

Among the 1,243 eligible residents, 1,004 were analysed to assess the antibody response after the first jab. Seven hundred and thirty-five residents had always tested negative for SARS-CoV-2 RNA by RT-PCR, 94 had tested positive 9 to 12 months before the vaccine and 176 had tested positive 3 to 7 months before. Among those 1,004 residents, 808 accepted the second jab and agreed to participate in the study (Figure 1).

N-Protein IgG levels 6 weeks after the first BNT162b2 jab

N-protein IgG were detectable ([?] 0.8 signal to cutoff ratio) in 42 (5.7%) of the 735 negative RT-PCR residents. These residents were then considered as immunised against SARS-CoV-2. Residents with an older infection were less frequently seropositive for N-protein IgG than residents with a newer infection (61.1% vs 83.7%, $p < .001$) (Table 1). The N-protein IgG median level was lower in residents with an older infection than in those with a newer infection ($p < 0.001$).

S-protein IgG levels 6 weeks after the first BNT162b2 jab

S-protein IgG was undetectable (< 50 AU/mL) in 41.7% of the RT-PCR-negative residents who had tested negative for N-protein IgG, in 11.9% of the residents with a negative RT-PCR but who were positive for N-protein IgG, in 2.1% of residents with an older infection, and in 4.6% with a newer infection (Table 1).

A low S-protein IgG level ([?] 1,050 UA/ml) was more often observed in residents without prior COVID-19 (negative for both RT-PCR and N-protein IgG tests) than in residents with a previous positive RT-PCR (91.3% vs 11.5%, $p < 0.001$). RT-PCR-positive residents with an older infection more often had a high S-protein IgG level than those with a newer infection (88.3% vs 84.0%, $p < 0.001$), and RT-PCR-negative residents tested positive for N-protein IgG (88.3% vs 73.8%, $p < 0.001$).

The predictive value of a high S-protein IgG level ([?] 4,160 UA/ml) by a prior RT-PCR in the last 3 to 12 months was high (positive predictive value of 85.5% [83.3%-87.7%] and negative predictive value of 94.0% [92.5%-95.5%]).

Link between S- and N-Protein IgG levels after the first BNT162b2 dose

Among the RT-PCR-positive residents, the median levels of S-protein IgG after vaccine were ranked in the following order: *i*) residents with a newer infection and no detectable N-protein IgG (median value of

230AU/mL), *ii*) residents with a newer infection and detectable N-protein IgG (median value of 20,685 AU/mL), *iii*) residents with an older infection and no detectable N-protein IgG (median value of 27,313 AU/mL), and *iv*) residents with an older infection and detectable N-protein IgG (median value of 40,000 AU/mL)(Table 2, Figure 2).

The predictive value of a high S-protein IgG level ([?] 4,160 UA/ ml) by N-protein IgG ([?] 0.8 signal to cutoff ratio) was high (positive predictive value of 92.5% [90.8%-94.2%] and negative predictive value of 93.0% [91.4%-94.6%]).

S-protein IgG levels 6 weeks after the second BNT162b2 jab

Six weeks after the second BNT162b2 jab, only around 3% of the 554 residents without prior COVID-19 had undetectable S-protein IgG levels (< 50 AU/mL) vs none of the residents with a prior positive PCR (Table 3). The prevalence of residents with low S-protein IgG levels ([?] 1,050 AU/mL) was significantly lower in residents without prior COVID-19 than in those with a positive PCR in the 3 to 12 last months (28.3% vs 2.7%, $p < 0.001$) (Figure 3). The median value of S-protein IgG in residents without prior COVID-19 after two vaccine doses was 10-fold lower than that in residents with prior COVID-19 after one dose (2,384 AU/mL vs 23,259 AU/mL)

Discussion

Recent studies conducted in small samples of immunocompetent adults showed higher levels of S-protein IgG after a single jab in individuals with prior COVID-19 than in those without prior COVID-19.^{5,23} Our pilot study with 136 residents was the only one to include NH residents.¹⁶ The current study is innovative because it includes a large sample of NH residents and evaluates the effect on antibody response of (i) the time elapsed since COVID-19 infection and vaccination, and (ii) the persistence of a natural immunity against SARS-CoV-2 N-protein at the time of the first vaccination. The large sample allows the assessment of the percentage of residents with undetectable S-protein IgG (< 50 AU/mL) or with low levels of S-protein ([?]1050 AU/mL) after two vaccine doses among those with or without prior COVID-19. It also enables the comparison of antibody response after one jab in residents without COVID-19 and after two doses in residents with prior COVID-19.

The present study shows that 3 weeks after the first vaccine dose: *i*) 41.7% of residents without prior COVID-19 have undetectable S-protein IgG and 91.3% have low levels of S-protein IgG, *ii*) only 3.8% of residents without prior COVID-19 have undetectable S-protein IgG and 11.5% have low levels of S-protein IgG, *iii*) among residents with prior COVID-19, those seropositive for N-protein at the time of the vaccine and those with an older infection (in the last 9 to 12 months) have the highest S-protein IgG levels, *iv*) the second jab significantly boosts the antibody response of residents without prior COVID-19 (since 6 weeks after the second dose, only 3.1% remain seronegative for S-protein IgG). However, *v*) 29.4% of residents without prior COVID-19 have low S-protein IgG levels, *vi*) in residents with prior COVID-19, the second dose leads to a reduced percentage of residents with low S-protein IgG levels (from 11.5% after the first dose to 2.7% after the second dose). The median value of S-protein IgG levels after one jab in residents with prior COVID-19 exceeds that of two doses in residents without prior COVID-19.

Findings of the present study in light of current publications on residents with prior COVID-19

Three weeks after a single dose of BNT162b2 vaccine, residents with an older infection were less often seropositive for N-protein IgG than those with a newer infection (61.1 % vs 85.7 %). This result accords with the time-related decline of natural SARS-CoV-2 antibodies observed not only in the general population with prior COVID-19²⁴ but also in NHs: a study showed that 91% of residents were still seropositive for N-protein 6 months after COVID-19, but with a decreasing antibody titer over time²⁵. The rapid decay of N-protein IgG in the 90 days after mild COVID-19 infection has suggested a short-lasting humoral immunity against SARS-CoV-2²⁶. The present study is quite reassuring as it indicates a natural immunity in 61% of NH residents 9 to 12 months after infection, which is in line with the results shown after other acute viral infections^{27,28}.

This study shows a link between N-protein IgG and S-protein IgG levels after the first jab. RT-PCR-positive residents who were seropositive for N-protein IgG more often exhibited a high S-protein IgG level after vaccination (over 96% of them) than seronegative residents with a positive RT-PCR from an older (80%) or newer (28.6%) infection. This result is in line with influenza for which pre-existing immunity in older individuals, contrary to immune-senescence or poor functional status, is a strong correlate of post-vaccination immunity^{29,30}.

It is reassuring to note that 80% of the RT-PCR-positive residents with an older infection but who do not have a persistent humoral immunity exhibit a high S-protein IgG level after the vaccine. This result suggests a long-term persistence of previously-generated memory B cells that can induce a rapid clonal expansion and terminal differentiation to produce high affinity anti-S-protein IgG after the vaccine³¹, as observed for common viruses³². In line with this hypothesis, a robust boosting after the first mRNA jab was found to strongly correlate with levels of pre-existing memory B cells in recovered individuals, identifying a key role for memory B cells in recall responses to SARS-CoV-2 antigens³³. Taken together, these results suggest that both natural humoral immunity (that appears to decrease over time) and cellular immunity (that seems to be maintained over time) may modulate the high level of SARS-CoV-2 S-protein IgG following a single BNT162b2 jab in nursing home residents having recovered from COVID-19. This highlights the utility of defining cellular responses in addition to serologies to predict the immune response to vaccine in this population. This result may be important since the role of pre-existing immunity was underscored during the 2009 A (H₁N₁) pandemic, when older adults were less severely affected than younger individuals³⁴.

The present study shows that NH residents with prior COVID-19 achieve much higher S-protein IgG levels after one single jab than in younger COVID-19 convalescents³⁵. It remains to be determined whether vaccinated residents with prior COVID-19 (with at least one jab) will be better protected against reinfection during outbreaks in NHs than unvaccinated residents with prior COVID-19⁴.

The persistence of N-protein IgG in 85.7% of residents with a newer infection (in the past 3 to 7 months) and the increase in S-protein level observed with increasing time between COVID-19 and vaccination both support the hypothesis that, in most residents, the vaccine should be administered at least 3 months after COVID-19 infection, even if reinfection is possible within those 3 months³⁶.

Strengths and limitations of the study

This study has several strengths. Our sample is probably representative of the population of NH residents in France since it was conducted on a large sample of residents who were tested across 22 NHs facing a COVID-19 outbreak. Ascertainment of positive and negative RT-PCR results is probably almost complete since all studied NHs followed the same regional Health Agency guidance published in March 2020^{2,3}. We used an automated quantitative assay to measure the RBD IgG level that correlates well with virus neutralisation^{37,38}. N-protein IgG measurement in all individuals allowed us to differentiate residents with SARS-CoV-2 immunisation while having repeated negative RT-PCR tests.

The main limitation of the study is the lack of clinical outcome. It remains indeed to demonstrate that a single BNT162b2 jab in residents having recovered from COVID-19 has the same efficacy in preventing reinfection as two doses. If the N-protein IgG level is associated with a substantially reduced risk of SARS-CoV-2 reinfection⁸, there is no available publication demonstrating a link between the S-protein IgG level obtained after the vaccine and the risk of incident SARS-CoV-2 infection and of symptomatic or severe COVID-19. It remains to be determined whether the thresholds we have chosen to define a low level ([?] 1050 AU/mL) or a high S-protein IgG after the vaccine ([?] 4,160 AU/mL) are effectively associated with an increased or decreased risk of developing SARS-CoV-2 infection.⁷ This is particularly important since serum neutralising activities against SARS-CoV-2 six months after COVID-19 hospitalisation remain significant for ancestral strains and for the D614G and B.1.16 variants but are weaker for the B.1.351 variant³⁹. It is therefore plausible that the post-vaccine S-protein IgG level necessary to obtain protection against new variants may be higher than previously defined⁴⁰.

Generalisability

Although this study has been carried out in one European region, its size allows generalisability in residents infected by the natural strain of the virus. New studies are needed to study the post-vaccine antibody response in residents infected by different variants.

Conclusions

Recent studies show that even if the vaccine is very effective in reducing severe forms of COVID-19 in nursing home residents, the vaccination, even complete, may not totally prevent the risk of SARS-CoV-2 outbreaks in nursing homes (Blain et al., submitted).⁴¹

This present study supports the hypothesis that *i*) most of the residents having recovered from COVID-19 (with a diagnosis using RT-PCR or N-protein IgG) may only require one single vaccine dose to achieve peak antibody and memory B cell responses. *ii*) However, the second dose reduces the prevalence of residents with low S-protein IgG levels ($[?]1050$ AU/mL)(from 11.5% to 2.7%). *iii*) For individuals whose infection history is unknown, measuring S-protein IgG antibody levels just before the second jab could be useful in determining whether a second dose is required. *iv*) Most of the individuals without prior COVID-19 have a low level of S-protein IgG 3 weeks after the first jab, further confirming the benefit to administer the second dose without delay to boost the antibody response. After two doses, around 30% of residents without prior COVID-19 have a low level of S-protein IgG. Whether these residents with low antibody response after the complete vaccine regimen are at higher risk of symptomatic COVID-19 and whether they may benefit from a third vaccine dose remains to be further investigated.

Additional studies are required to demonstrate whether the measure of S-protein IgG after the vaccination should help to personalise the vaccine schedules and reduce secondary effects related to possible reactogenicity⁴². Further studies are necessary also to determine whether the present results obtained in residents infected by the natural strain of the virus may be replicated in residents infected by the new variants^{43,44}.

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Conflicts of Interest:

The authors declare no conflicts of interest/competing interests

Abbreviations

N-protein IgG: nucleocapsid (N) IgG protein

NH: Nursing home

RT-PCR: real-time reverse-transcriptase polymerase chain reaction for SARS-CoV-2

S-protein IgG: SARS-CoV-2 spike IgG

SARS-CoV-2: Severe Acute Respiratory Syndrome Coronavirus-2

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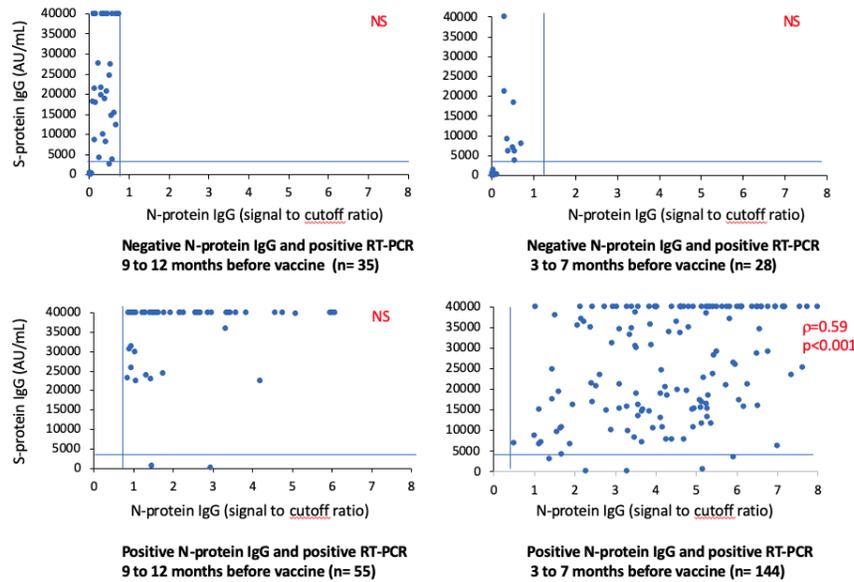
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Figure 1. Flow Diagram of the sample of residents from 22 nursing homes having faced a COVID-19 outbreak between March and December 2020.

Figure 2. Correlation between N- and S-protein IgG depending on the time of the COVID-19 infection 3 weeks after the first jab



Legend: N-protein IgG levels: positive value when [?] 0.8 signal to cutoff ratio (vertical bars); S-protein IgG levels: high values when [?] 4160 arbitrary unit (AU)/mL (horizontal bar)

Figure 3: S-protein IgG levels in residents with or without prior COVID-19 3 weeks after the first jab and 6 weeks after the second

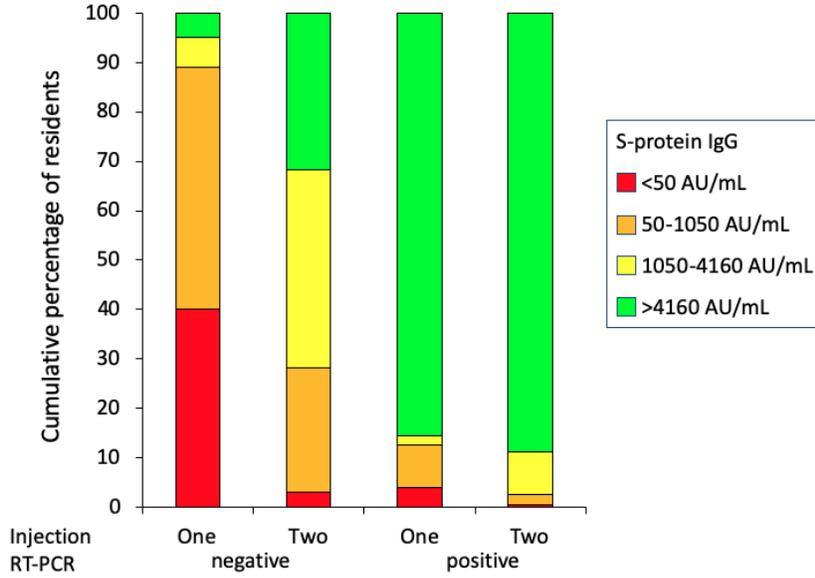


Table 1: Demographic characteristics and N-Protein IgG and S-Protein levels of the whole sample of nursing home residents 3 weeks after the first jab

| | Repeated negative RT-PCR in the last 12 months (n=735) | Repeated negative RT-PCR in the last 12 months (n=735) | Positive RT-PCR (n=270) | Positive RT-PCR (n=270) | Positive RT-PCR (n=270) | <i>P</i> value | |
|------------------------------------------------------------------------|--------------------------------------------------------|--------------------------------------------------------|--------------------------------------------|----------------------------------------|----------------------------------------|-----------------------------------------------|-----------------------------------------------|
| | Negative N-Protein IgG ^a (n=693) | Positive N-Protein IgG ^b (n=42) | Positive N-Protein IgG ^b (n=42) | 9 to 12 months ago ^c (n=94) | 3 to 7 months ago ^d (n=176) | | |
| Sex Female (%) | 484 (69.8) | 35 (83.3) | 35 (83.3) | 66 (70.2) | 139 (78.0) | .03 | .03 |
| Male (%) | 209 (30.2) | 7 (16.7) | 7 (16.7) | 28 (29.8) | 37 (21.0) | | |
| Age, mean (SD), year | 86.2 (0.0) | 85.3 (7.0) | 85.3 (7.0) | 86.6 (9.2) | 87.8 (8.2) | .05 | .05 |
| SARS-CoV-2 N-Protein IgG level [?] 0.8 signal to cutoff ratio, No. (%) | 0 | 42 (100) | 42 (100) | 55 (61.1) | 144 (83.7) | <.001 between ^c and ^d | <.001 between ^c and ^d |
| SARS-CoV-2 N-Protein IgG level, median (IQR) [range], AU/mL | 0 | 3.4 (0.8;7.3), [0.8-7.3] | 3.4 (0.8;7.3), [0.8-7.3] | 1.0 (0.4;2.6) [0.02-6.1] | 3.91 (1.9;5.3) [0.01-8] | <.001 between ^c and ^{b,d} | <.001 between ^c and ^{b,d} |

| | Repeated negative RT-PCR in the last 12 months (n=735) | Repeated negative RT-PCR in the last 12 months (n=735) | Positive RT-PCR (n=270) | Positive RT-PCR (n=270) | Positive RT-PCR (n=270) | <i>P</i> value | |
|----------------------------------------------------------|--------------------------------------------------------|--------------------------------------------------------|----------------------------|---------------------------------|--------------------------------|-------------------------------------------------|-------------------------------------------------|
| SARS-CoV-2 S-protein IgG level | | | | | | | |
| 0-50 AU/mL | 288 (41.7) | 5 (11.9) | 5 (11.9) | 8 (4.6) | 2 (2.1) | | |
| 50-1050 AU/mL | 343 (49.6) | 5 (11.9) | 5 (11.9) | 15 (8.6) | 6 (6.4) | | |
| 1050-4160 AU/mL | 47 (6.8) | 1 (2.4) | 1 (2.4) | 5 (2.8) | 3 (3.2) | | |
| [?] 4160 AU/mL | 13 (1.9) | 31 (73.8) | 31 (73.8) | 147 (84.0) | 83 (88.3) | | |
| SARS-CoV-2 S-protein level median (Q1-Q3) [range], AU/mL | 82 (16-296)[0-40,000] | 21,736 (16,229) [0-40,000] | 21,736 (16,229) [0-40,000] | 40,000 (9,146-40,000) [6-14142] | 20,796 (4157;40000) [1-40,000] | <.001 between ^a and ^{b,c,d} | <.001 between ^a and ^{b,c,d} |
| | | | | | | <.001 between ^c and ^{b,d} | <.001 between ^c and ^{b,d} |

Table 2: S-Protein IgG levels in RT-PCR-positive residents 3 weeks after the first jab

| | Negative N-Protein IgG in the last 9 to 12 months ^a | Positive N-Protein IgG in the last 9 to 12 months ^b | Negative N-Protein IgG in the last 3 to 7 months ^c | Positive N-Protein IgG in the last 3 to 7 months ^d | <i>P</i> value |
|--------------------------------|----------------------------------------------------------------|----------------------------------------------------------------|---------------------------------------------------------------|---------------------------------------------------------------|---------------------------------------------------|
| IgG level [?] 50 AU/ml No. (%) | 39 (97.1) | 55 (100.0) | 32 (75.0) | 144 (100.0) | p<0.001 between ^c and ^{a,b,d} |
| median level [range], AU/ml | 20,685 [13,-40,000] | 40,000 [67-40,000] | 230 [1- 40,000] | 27313 [210-40,000] | p<0.001 between ^c and ^{a,b,d} |

Abbreviations: AU, arbitrary units; IQR, interquartile range

| | Repeated negative RT-PCR in the last 12 months (n=589) | Repeated negative RT-PCR in the last 12 months (n=589) | Positive RT-PCR (n=222) | Positive RT-PCR (n=222) | <i>P</i> value |
|--|--------------------------------------------------------|--------------------------------------------------------|----------------------------------------|----------------------------------------|----------------|
| | Negative N-Protein IgG ^a (n= 554) | Positive N-Protein IgG ^b (n=35) | 9 to 12 months ago ^c (n=72) | 3 to 7 months ago ^d (n=150) | |

| | Repeated negative RT-PCR in the last 12 months (n=589) | Repeated negative RT-PCR in the last 12 months (n=589) | Positive RT-PCR (n=222) | Positive RT-PCR (n=222) | <i>P</i> value |
|-------------------------------------------------------------------|--------------------------------------------------------------------|--------------------------------------------------------------------|----------------------------------|-----------------------------------|--------------------------------------------------------------------------------------------------------|
| Sex Female (%) | 387 (69.9) | 32 (91.4) | 52 (72.2) | 121 (80.7) | <.01 |
| Male (%) | 167 (30.1) | 3 (8.6) | 20 (27.8) | 29 (19.3) | |
| Age, mean (SD), year | 86.2 (9.0) | 85.6 (7.2) | 87.4 (8.6) | 88.0 (8.5) | NS |
| SARS-CoV-2 S-protein IgG level | | | | | |
| 0-50 AU/mL | 17 (3.1) | 1 (2.9) | 0 (0) | 1 (0.7) | |
| 50-1050 AU/mL | 146 (26.3) | 2 (5.7) | 0 (0) | 5 (3.3) | |
| 1050-4160 AU/mL | 231 (41.7) | 5 (14.3) | 6 (8.3) | 12 (8.0) | |
| [?] 4160 AU/mL | 160 (28.9) | 27 (77.1) | 66 (91.7) | 132 (88.0) | |
| SARS-CoV-2 S-protein level median (Q1-Q3) [range], AU/mL | 2384 (827; 4902)[0-40,000] | 18239 (5876; 38418) [0; 40000][0-40,000] | 39525 (19601; 4000) [6-14142] | 28863 (12083; 40000)[1-40,000] | <.001 between ^a and ^{b,c,d} <.001 between ^c and ^{b,d} |

Table 3: Demographic characteristics and N-Protein IgG and S-Protein levels of the whole sample of nursing home residents after the second jab