nature

https://doi.org/10.1038/s41586-022-04830-x

Accelerated Article Preview

Omicron infection enhances Delta antibody immunity in vaccinated persons

Received: 27 January 2022

Accepted: 4 May 2022

Accelerated Article Preview

Cite this article as: Khan, K. et al. Omicron infection enhances Delta antibody immunity in vaccinated persons. *Nature* https://doi.org/10.1038/s41586-022-04830-x (2022)

Khadija Khan, Farina Karim, Sandile Cele, Kajal Reedoy, James Emmanuel San, Gila Lustig, Houriiyah Tegally, Yuval Rosenberg, Mallory Bernstein, Zesuliwe Jule, Yashica Ganga, Nokuthula Ngcobo, Matilda Mazibuko, Ntombifuthi Mthabela, Zoey Mhlane, Nikiwe Mbatha, Yoliswa Miya, Jennifer Giandhari, Yajna Ramphal, Taryn Naidoo, Aida Sivro, Natasha Samsunder, Ayesha B. M. Kharsany, Daniel Amoako, Jinal N. Bhiman, Nithendra Manickchund, Quarraisha Abdool Karim, Nombulelo Magula, Salim S. Abdool Karim, Glenda Gray, Willem Hanekom, Anne von Gottberg, COMMIT-KZN Team*, Ron Milo, Bernadett I. Gosnell, Richard J. Lessells, Penny L. Moore, Tulio de Olveira, Mahomed-Yunus S. Moosa & Alex Sigal

Omicron infection enhances Delta antibody immunity in vaccinated persons

Khadija Khan[1,2#], Farina Karim[1,2#], Sandile Cele[1,2], Kajal Reedoy[1], James Emmanuel San[3], Gila Lustig[4], Houriiyah Tegally[3,5], Yuval Rosenberg[6], Mallory Bernstein[1], Zesuliwe Jule[1], Yashica Ganga[1], Nokuthula Ngcobo[1], Matilda Mazibuko[1], Ntombifuthi Mthabela[1], Zoey Mhlane[1], Nikiwe Mbatha[1], Yoliswa Miya[1], Jennifer Giandhari[3], Yajna Ramphal[3], Taryn Naidoo[1], Aida Sivro[4,7], Natasha Samsunder[4], Ayesha BM. Kharsany[4], Daniel Amoako[8], Jinal N. Bhiman [8], Nithendra Manickchund[9], Quarraisha Abdool Karim[4,11], Nombulelo Magula[10], Salim S. Abdool Karim[4,11], Glenda Gray[12], Willem Hanekom[1,13], Anne von Gottberg[13,14], COMMIT-KZN Team[§], Ron Milo[6], Bernadett I. Gosnell[9], Richard J. Lessells[3,4], Penny L. Moore[4,13,14,15], Tulio de Oliveira[3,4,5,16], Mahomed-Yunus S. Moosa9, Alex Sigal[1,2,4,17*]

[1]Africa Health Research Institute, Durban, South Africa. [2]School of Laboratory Medicine and Medical Sciences, University of KwaZulu-Natal, Durban, South Africa. [3]KwaZulu-Natal Research Innovation and Sequencing Platform, Durban, South Africa. [4]Centre for the AIDS Programme of Research in South Africa, Durban, South Africa. [5]Centre for Epidemic Response and Innovation, School of Data Science and Computational Thinking, Stellenbosch University, Stellenbosch, South Africa.[6]Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, Israel. [7]Department of Medical Microbiology, University of KwaZulu-Natal, Durban, South Africa. [8]National Institute for Communicable Diseases of the National Health Laboratory Service, Johannesburg, South Africa. [9]Department of Infectious Diseases, Nelson R. Mandela School of Clinical Medicine, University of KwaZulu-Natal, Durban, South Africa. [10]Department of Internal Medicine, Nelson R. Mandela School of Medicine. University of Kwa-Zulu Natal. [11]Department of Epidemiology, Mailman School of Public Health, Columbia University, New York, NY, United States. [12]South African Medical Research Council, Cape Town, South Africa. [13]Division of Infection and Immunity, University College London, London, UK. [14]SAMRC Antibody Immunity Research Unit, School of Pathology, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa. [15]Institute of Infectious Disease and Molecular Medicine, University of Cape Town, Cape Town, South Africa.[16]Department of Global Health, University of Washington, Seattle, USA.[17]Max Planck Institute for Infection Biology, Berlin, Germany.

[#] Equal contribution.

* Corresponding author. Email: alex.sigal@ahri.org

The extent to which Omicron infection[1-9], with or without previous vaccination, elicits protection against the previously dominant Delta (B.1.617.2) variant is unclear. We measured SARS-CoV-2 variant neutralization capacity in 39 Omicron sub-lineage BA.1 infected individuals in South Africa starting at a median of 6 (IQR 3-9) days post-symptoms onset and continuing until a last follow-up sample a median of 23 (IQR 19-27) days post-symptoms to allow BA.1 elicited neutralizing immunity time to develop. Fifteen participants were vaccinated with Pfizer-BNT162b2 or J&J-Ad26.CoV2.S and had BA.1 breakthrough infections, and 24 were unvaccinated. BA.1 neutralization increased from a geometric mean titer (GMT) $FRNT_{50}$ of 42 at enrollment to 575 at the last follow-up time-point (13.6-fold) in vaccinated and from 46 to 272 (6.0-fold) in unvaccinated participants. Delta virus neutralization also increased, from 192 to 1091 (5.7-fold) in vaccinated and 28 to 91 (3.0-fold) in unvaccinated participants. At the last time-point, unvaccinated BA.1 infected individuals had 2.2-fold lower BA.1 neutralization, 12.0-fold lower Delta neutralization, 9.6-fold lower Beta variant neutralization, 17.9-fold lower ancestral virus neutralization, and 4.8-fold lower Omicron sub-lineage BA.2 neutralization relative to vaccinated, with low absolute levels of neutralization for the non-BA.1 viruses. These results indicate that vaccination combined with Omicron/BA.1 infection hybrid immunity should be protective against Delta and other variants. In contrast, infection with Omicron/BA.1 alone offers limited cross-protection despite moderate enhancement.

The Omicron variant of SARS-CoV-2, first identified in November 2021 in South Africa and Botswana[10], has been shown by us[1] and others[2-9] to have extensive but incomplete escape from neutralizing immunity elicited by vaccines and previous infection, with boosted individuals showing better neutralization. In South Africa, Omicron infections led to a lower incidence of severe disease relative to other variants[11,12], although this can be at least partly explained by pre-existing immunity[13]. The first Omicron sub-lineage to appear was BA.1, which was supplanted by the BA.2 sub-lineage in many countries[14].

How Omicron/BA.1 infection will interact with vaccination to protect against the previously dominant Delta variant, emerging variants such as BA.2, and other variants, is still unclear. We isolated live Omicron/BA.1, Omicron/BA.2, ancestral, Beta, and Delta viruses and neutralized viruses with plasma from participants enrolled and longitudinally sampled during the Omicron/BA.1 infection wave in South Africa, with all participants having a confirmed diagnosis of SARS-CoV-2 by qPCR. To quantify neutralization, we used a live virus neutralization assay and calculated the focus reduction neutralization test ($FRNT_{50}$) value, the inverse of the plasma dilution required for 50% neutralization, as measured by the reduction in the number of infection foci. We enrolled 41 participants who reported symptoms from late November 2021 to January 2022. We successfully sequenced the infecting virus in 26 participants and all sequences corresponded to Omicron/BA.1 (Extended Data Table 1). Two participants had advanced HIV disease based on a low CD4 count (<200 cells/microliter throughout the study) and unsuppressed HIV infection, and we excluded these participants based on our previous data showing an atypical response to SARS-CoV-2 in advanced HIV disease[15]. Extended Data Table 2 summarizes the characteristics of the remaining 39 participants.

Twenty seven out of 39 participants were admitted to hospital because of Covid-19 symptoms. Seven required supplemental oxygen and one died. Fifteen participants were vaccinated and had a breakthrough Omicron/BA.1 infection. The median time post-vaccination was 139 days (IQR 120-178), a time interval which would predict considerable waning of the vaccine elicited immune response[16] which may have contributed to the breakthrough infections. Eight participants were vaccinated with two doses of Pfizer-BNT162b2 and 7 with Johnson and Johnson Ad26.CoV2.S, with one Ad26.CoV2.S vaccinee boosted with a second Ad26.CoV2.S dose (Extended Data Table 1). The length of hospital stay was shorter in the vaccinated (3.5 days) relative to unvaccinated participants (8 days, Extended Data Table 2). Three participants self-reported having a previous SARS-CoV-2 infection (Extended Data Table 1).

Participants were sampled at enrollment at a median of 6 days (interquartile range 3-9 days) post-symptom onset, and again at weekly follow-up visits which were attended as practicable because of the Christmas holidays in South Africa. The last follow-up visit was a median of 23 days (interquartile range 19-27 days) post-symptom onset (Extended Data Table 1). Examining neutralization at all available timepoints per study participant showed that neutralization of the Omicron/BA.1 variant increased substantially in most participants from enrollment to time of last follow-up (Extended Data Fig 1), consistent with developing a neutralizing antibody response to Omicron/BA.1 infection. We therefore analyzed neutralization at enrollment (baseline for the study) and the last follow-up visit to quantify the increase in neutralization capacity post-Omicron infection.

We observed that Omicron/BA.1 neutralization increased in vaccinated individuals from a low geometric mean titer (GMT) $FRNT_{50}$ of 42 at the enrollment visit to 575 at the last follow-up visit about 2 to 3 weeks later, a 13.6-fold change (95% CI 3.7-50.2, Fig 1a). The samples from unvaccinated participants neutralized at study baseline at a similar starting level of 46 and reached a final level of 272 at last follow-up, a 6.0-fold increase (95% CI 2.2-16.1, Fig 1b). Neutralization of Delta virus also increased during this period. At enrollment, neutralization capacity against Delta virus was 192 and reached a final level of 1091 at last follow-up in vaccinated participants, a 5.7-fold increase (95% CI

1.7-18.4, Fig 1c). Unvaccinated participants had lower Delta neutralization at baseline with Delta virus $FRNT_{50}$ = 28, and reached $FRNT_{50}$ = 91, a 3.2-fold increase (95% CI 1.3-8.1, Fig 1d).

We next compared Omicron/BA.1 to Omicron/BA.2, Delta, Beta[17] and ancestral virus neutralization at the last available follow-up visit in three sets of paired experiments, each comparing either Omicron/BA.2, Delta, or ancestral and Beta virus neutralization to Omicron/BA.1 neutralization. The range of Omicron/BA.1 neutralization shown in Fig 2a for different experiments ($FRNT_{50}$ = 516 to 646 for vaccinated samples and 266 to 271 for unvaccinated samples) is the result of experimental variation. BA.2 neutralization was moderately and not significantly lower relative to BA.1 in both vaccinated and unvaccinated participants. Testing only participants with sequence confirmed Omicron BA.1 infection gave a similar result (Extended Data Fig 2). The trend for the other variants and ancestral virus was that neutralization was higher relative to Omicron/BA.1 in vaccinated participants but lower relative to Omicron/BA.1 in unvaccinated participants, although the differences were mostly not significant (Fig 2a). Because of the relatively moderate fold-change, higher participant numbers would likely be required make the trends statistically significant.

The comparison of the other variants to Omicron/BA.1 does not indicate the differences in neutralization capacity elicited by Omicron/BA.1 in vaccinated versus unvaccinated participants, since Omicron/BA.1 neutralization capacity is itself different between the vaccinated and unvaccinated groups. We therefore compared neutralization of each variant between the vaccinated and unvaccinated groups at the last timepoint directly (Fig 2b). The smallest difference between vaccinated and unvaccinated participants was in neutralization of Omicron/BA.1, the infecting variant, with the vaccinated participants showing 2.2-fold higher neutralization. For the other variants, cross-neutralization was higher in vaccinated participants by a factor of 4.8-fold for Omicron/BA.2, 9.6-fold for Beta, 12.0-fold for Delta, and 17.9-fold for ancestral (Fig 2b). All differences were significant, and the 95% confidence intervals for the GMT $FRNT_{50}$ of vaccinated and unvaccinated participants did not overlap for BA.2, Beta, Delta, or ancestral virus neutralization (Fig 2b). For the unvaccinated participants, absolute neutralization capacity of BA.2, Beta, Delta, and ancestral virus was low[18], with GMT $FRNT_{50}$ being about or below $FRNT_{50}$ = 100 (Fig 2b).

We also tested neutralization of Omicron/BA.1 by Delta variant elicited immunity. We collected 18 plasma samples from 14 participants (including pre- and post-vaccination samples from 4 participants) previously infected in the Delta variant wave in South Africa, eight of whom were vaccinated either before or after infection (Extended Data Table 3). Confirming previously reported results[19], we observed similar extensive escape of Omicron/BA.1 from Delta elicited immunity across all samples tested, manifested as a 22.5-fold decrease (95% CI 14.4-35.0) in Omicron/BA.1 neutralization compared to Delta virus neutralization (Fig 3).

The large fold-drop in Delta infection elicited neutralization capacity against Omicron/BA.1 contrasts with the moderate and non-significant fold-drops, or even fold-increases, in neutralization of other variants relative to Omicron/BA.1 in Omicron/BA.1 infected individuals. However, in unvaccinated individuals, even though fold-drops in neutralization were moderate and non-significant, the absolute levels of neutralization of the other variants, and of Omicron/BA.1 itself, were low and on a similar scale to the cross-neutralization capacity against Omicron in Delta infection elicited immunity. This is consistent with other recently reported results[20], and possibly indicates that Omicron is poorly immunogenic. In agreement with recent reports[21,22], we observed moderately and non-significantly lower neutralization of BA.2 by BA.1 elicited immunity. The results explain epidemiological observations showing that Omicron/BA.2 re-infection is relatively rare soon after Omicron/BA.1 infection[23,24].

Our results may be supportive of a scenario where hybrid immunity formed by Omicron infection combined with vaccination protects as well or better against re-infection with current variants such as Delta relative to re-infection with Omicron itself. In contrast, unvaccinated participants infected

with Omicron/BA.1 have low absolute neutralization capacity of Omicron/BA.2, Beta, Delta, and ancestral virus, and neutralizing immunity to these variants is dramatically lower relative to the BA.1 infected vaccinated participants.

Limitations of this study include heterogeneity in participant immune history, including two vaccination types and one boost. Based on the high seroprevalence observed in South Africa[25,26], some participants may also have had unreported previous infection. However, including two vaccine types did not mask the differences between vaccinated and unvaccinated participants, and the low levels of neutralization in unvaccinated participants against ancestral, Beta, and Delta viruses (the dominant strains in the preceding South African infection waves) supports the notion that these participants were either not previously infected, or that immunity has waned completely. Participants were also mostly hospitalized, which may not be typical of Omicron infection[13,27]. Increased disease severity has been shown to lead to higher anti-SARS-CoV-2 antibody titers[28]. This should help in the detection of the neutralization response, but whether it would affect the trend we observed is unclear. Omicron infection is unlikely benign to the extent where hospitalization is an outlier outcome: In the USA, the number of Covid-19 cases who died in the Omicron wave was similar to the number who died in the Delta wave[27]. Neutralizing immunity may have increased further in some participants had we sampled later: Neutralizing capacity did not plateau at the last time-point in 8 of 24 (33%) of unvaccinated participants (participants 10, 14, 21, 27, 30, 31, 34, and 38, Extended Data Fig 1) and 6 of the 15 (40%) of vaccinated participants (participants 4, 6, 15, 16, 25, and 26). Therefore, the temporal dynamics give no clear indication that the immunity in the unvaccinated participants was delayed and would have reached similar levels to that of vaccinated participants if sampled later. However, the consequences of waning immunity several months post-Omicron infection should be investigated.

The gap in immunity between unvaccinated Omicron/BA.1 infected individuals and those vaccinated with BA.1 breakthrough infection is concerning. Especially as immunity wanes, unvaccinated individuals post-Omicron infection are likely to have poor cross-protection against existing and possibly emerging SARS-CoV-2 variants, despite acquiring some neutralizing immunity to the infecting Omicron variant. The implication may be that Omicron infection alone is not sufficient for protection and vaccination should be administered even in areas with high prevalence of Omicron infection to protect against other variants.

References

1 Cele, S. *et al.* Omicron extensively but incompletely escapes Pfizer BNT162b2 neutralization. *Nature*, doi:10.1038/s41586-021-04387-1 (2021).

2 Andrews, N. *et al.* Covid-19 Vaccine Effectiveness against the Omicron (B.1.1.529) Variant. *New England Journal of Medicine*, doi:10.1056/NEJMoa2119451 (2022).

3 Cao, Y. *et al.* Omicron escapes the majority of existing SARS-CoV-2 neutralizing antibodies. *Nature* **602**, 657-663, doi:10.1038/s41586-021-04385-3 (2022).

4 Dejnirattisai, W. *et al.* SARS-CoV-2 Omicron-B.1.1.529 leads to widespread escape from neutralizing antibody responses. *Cell*, doi:10.1016/j.cell.2021.12.046 (2022).

5 Dejnirattisai, W. *et al.* Reduced neutralisation of SARS-CoV-2 omicron B.1.1.529 variant by post-immunisation serum. *Lancet* **399**, 234-236, doi:10.1016/s0140-6736(21)02844-0 (2022).

6 Garcia-Beltran, W. F. *et al.* mRNA-based COVID-19 vaccine boosters induce neutralizing immunity against SARS-CoV-2 Omicron variant. *Cell* **185**, 457-466.e454, doi:10.1016/j.cell.2021.12.033 (2022).

7 Lu, L. *et al.* Neutralization of SARS-CoV-2 Omicron variant by sera from BNT162b2 or Coronavac vaccine recipients. *Clin Infect Dis*, doi:10.1093/cid/ciab1041 (2021).

8 Planas, D. *et al.* Considerable escape of SARS-CoV-2 Omicron to antibody neutralization. *Nature* **602**, 671-675, doi:10.1038/s41586-021-04389-z (2022).

9 VanBlargan, L. A. *et al.* An infectious SARS-CoV-2 B.1.1.529 Omicron virus escapes neutralization by therapeutic monoclonal antibodies. *Nature Medicine*, doi:10.1038/s41591-021-01678-y (2022).

10 Viana, R. *et al.* Rapid epidemic expansion of the SARS-CoV-2 Omicron variant in southern Africa. *Nature*, doi:doi: https://doi.org/10.1038/d41586-021-03832-5 (2021).

11 Davies, M.-A. *et al.* Outcomes of laboratory-confirmed SARS-CoV-2 infection in the Omicron-driven fourth wave compared with previous waves in the Western Cape Province, South Africa. *medRxiv*, 2022.2001.2012.22269148, doi:10.1101/2022.01.12.22269148 (2022).

12 Wolter, N. *et al.* Early assessment of the clinical severity of the SARS-CoV-2 omicron variant in South Africa: a data linkage study. *Lancet* **399**, 437-446, doi:10.1016/s0140-6736(22)00017-4 (2022).

13 Sigal, A. Milder disease with Omicron: is it the virus or the pre-existing immunity? *Nature Reviews Immunology* **22**, 69-71, doi:10.1038/s41577-022-00678-4 (2022).

14 Lyngse, F. P. *et al.* Transmission of SARS-CoV-2 Omicron VOC subvariants BA.1 and BA.2: Evidence from Danish Households. *medRxiv*, 2022.2001.2028.22270044, doi:10.1101/2022.01.28.22270044 (2022).

15 Cele, S. *et al.* SARS-CoV-2 prolonged infection during advanced HIV disease evolves extensive immune escape. *Cell Host & Microbe*, doi:10.1016/j.chom.2022.01.005 (2022).

16 Goldberg, Y. *et al.* Waning Immunity after the BNT162b2 Vaccine in Israel. *N Engl J Med* **385**, e85, doi:10.1056/NEJMoa2114228 (2021).

17 Cele, S. *et al.* Escape of SARS-CoV-2 501Y.V2 from neutralization by convalescent plasma. *Nature* **593**, 142-146, doi:10.1038/s41586-021-03471-w (2021).

18 Khoury, D. S. *et al.* Neutralizing antibody levels are highly predictive of immune protection from symptomatic SARS-CoV-2 infection. *Nature medicine*, 1-7 (2021).

19 Rössler, A., Riepler, L., Bante, D., von Laer, D. & Kimpel, J. SARS-CoV-2 Omicron Variant Neutralization in Serum from Vaccinated and Convalescent Persons. *N Engl J Med*, doi:10.1056/NEJMc2119236 (2022).

20 Servellita, V. *et al.* Neutralizing immunity in vaccine breakthrough infections from the SARS-CoV-2 Omicron and Delta variants. *Cell*, doi:10.1016/j.cell.2022.03.019.

21 Yu, J. *et al.* Comparable Neutralization of the SARS-CoV-2 Omicron BA.1 and BA.2 Variants. *medRxiv*, 2022.2002.2006.22270533, doi:10.1101/2022.02.06.22270533 (2022).

22 Richardson, S. I. *et al.* SARS-CoV-2 Omicron triggers cross-reactive neutralization and Fc effector functions in previously vaccinated, but not unvaccinated individuals. *medRxiv*, 2022.2002.2010.22270789, doi:10.1101/2022.02.10.22270789 (2022).

23 Chemaitelly, H. *et al.* Protection of Omicron sub-lineage infection against reinfection with another Omicron sub-lineage. *medRxiv*, 2022.2002.2024.22271440, doi:10.1101/2022.02.24.22271440 (2022).

24 Stegger, M. *et al.* Occurrence and significance of Omicron BA.1 infection followed by BA.2 reinfection. *medRxiv*, 2022.2002.2019.22271112, doi:10.1101/2022.02.19.22271112 (2022).

25 Khan, K. *et al.* Immunogenicity of SARS-CoV-2 infection and Ad26.CoV2.S vaccination in people living with HIV. *Clin Infect Dis*, doi:10.1093/cid/ciab1008 (2021).

26 Madhi, S. A. *et al.* Population Immunity and Covid-19 Severity with Omicron Variant in South Africa. *New England Journal of Medicine*, doi:10.1056/NEJMoa2119658 (2022).

27 Sigal, A., Milo, R. & Jassat, W. Estimating disease severity of Omicron and Delta SARS-CoV-2 infections. *Nature Reviews Immunology*, doi:10.1038/s41577-022-00720-5 (2022).

28 Smith, M. *et al.* Age, Disease Severity and Ethnicity Influence Humoral Responses in a Multi-Ethnic COVID-19 Cohort. *Viruses* **13**, doi:10.3390/v13050786 (2021).

Materials and methods

Informed consent and ethical statement

Blood samples and the Delta isolate were obtained after written informed consent from adults with PCR-confirmed SARS-CoV-2 infection who were enrolled in a prospective cohort study at the Africa Health Research Institute approved by the Biomedical Research Ethics Committee at the University of KwaZulu–Natal (reference BREC/00001275/2020). The Omicron/BA.1 was isolated from a residual swab sample with SARS-CoV-2 isolation from the sample approved by the University of the Witwatersrand Human Research Ethics Committee (HREC) (ref. M210752). The sample to isolate Omicron/BA.2 was collected after written informed consent as part of the "COVID-19 transmission and natural history in KwaZulu-Natal, South Africa: Epidemiological Investigation to Guide Prevention and Clinical Care" Centre for the AIDS Programme of Research in South Africa (CAPRISA) study and approved by the Biomedical Research Ethics Committee at the University of KwaZulu–Natal (reference BREC/00001195/2020, BREC/00003106/2021).

Data availability statement

Sequences of outgrown Omicron sub-lineages have been deposited in GISAID with accession EPI_ISL_7886688 (Omicron/BA.1) and EPI_ISL_9082893 (Omicron/BA.2). Delta, Beta, and ancestral SARS-CoV-2 isolates have been previously described[15]. Raw images of the data are available upon reasonable request.

Reagent availability statement

Virus isolates and cell line are available from the corresponding author. A Biosafety Level 3 facility is required for laboratories receiving live SARS-CoV-2.

Competing interest statement

Salim S. Abdool Karim is a member in the COVID advisory panel for Emerging Markets at Pfizer. The authors declare no other competing interests.

Author contributions

AS, KK, and FK conceived the study and designed the study and experiments. AvG, QAK, SSAK, GL, ASi, and NS identified and provided virus samples. M-YSM, FK, BIG, MB, KK, TN, MM, NM, ZM, NN, YM, NMb, NMa, NMag, ZJ, KR, and YG set up and managed the cohort and cohort data. KK, ZJ, KR, SC, HT, JES, YG, JG, YR, AK, DA, and JB performed experiments and sequence analysis with input from AS, TdO, RJL, and JNB. AS, KK, FK, RM, and YR interpreted data with input from M-YSM, GG, SSAK, WH, TdO, NMag, RJL, PLM. AS, KK, GL, FK, and MB prepared the manuscript with input from all authors.

Whole-genome sequencing, genome assembly and phylogenetic analysis

RNA was extracted on an automated Chemagic 360 instrument, using the CMG-1049 kit (Perkin Elmer, Hamburg, Germany). The RNA was stored at −80◦C prior to use. Libraries for whole genome sequencing were prepared using either the Oxford Nanopore Midnight protocol with Rapid Barcoding or the Illumina COVIDseq Assay. For the Illumina COVIDseq assay, the libraries were prepared according to the manufacturer's protocol. Briefly, amplicons were tagmented, followed by indexing using the Nextera UD Indexes Set A. Sequencing libraries were pooled, normalized to 4 nM and denatured with 0.2 N sodium acetate. An 8 pM sample library was spiked with 1% PhiX (PhiX Control v3 adaptor-ligated library used as a control). We sequenced libraries on a 500-cycle v2 MiSeq Reagent Kit on the Illumina MiSeq instrument (Illumina). On the Illumina NextSeq 550 instrument, sequencing was performed using the Illumina COVIDSeq protocol (Illumina Inc, USA), an amplicon-based next-generation sequencing approach. The first strand synthesis was carried using random hexamers

primers from Illumina and the synthesized cDNA underwent two separate multiplex PCR reactions. The pooled PCR amplified products were processed for tagmentation and adapter ligation using IDT for Illumina Nextera UD Indexes. Further enrichment and cleanup was performed as per protocols provided by the manufacturer (Illumina Inc). Pooled samples were quantified using Qubit 3.0 or 4.0 fluorometer (Invitrogen Inc.) using the Qubit dsDNA High Sensitivity assay according to manufacturer's instructions. The fragment sizes were analyzed using TapeStation 4200 (Invitrogen). The pooled libraries were further normalized to 4nM concentration and 25 µL of each normalized pool containing unique index adapter sets were combined in a new tube. The final library pool was denatured and neutralized with 0.2N sodium hydroxide and 200 mM Tris-HCL (pH7), respectively. 1.5 pM sample library was spiked with 2% PhiX. Libraries were loaded onto a 300-cycle NextSeq 500/550 HighOutput Kit v2 and run on the Illumina NextSeq 550 instrument (Illumina, San Diego, CA, USA). For Oxford Nanopore sequencing, the Midnight primer kit was used as described by Freed and Silander55. cDNA synthesis was performed on the extracted RNA using LunaScript RT mastermix (New England BioLabs) followed by gene-specific multiplex PCR using the Midnight Primer pools which produce 1200bp amplicons which overlap to cover the 30-kb SARS-CoV-2 genome. Amplicons from each pool were pooled and used neat for barcoding with the Oxford Nanopore Rapid Barcoding kit as per the manufacturer's protocol. Barcoded samples were pooled and bead-purified. After the bead clean-up, the library was loaded on a prepared R9.4.1 flow-cell. A GridION X5 or MinION sequencing run was initiated using MinKNOW software with the base-call setting switched off. We assembled paired-end and nanopore.fastq reads using Genome Detective 1.132 (https://www.genomedetective.com) which was updated for the accurate assembly and variant calling of tiled primer amplicon Illumina or Oxford Nanopore reads, and the Coronavirus Typing Tool56. For Illumina assembly, GATK HaploTypeCaller --min-pruning 0 argument was added to increase mutation calling sensitivity near sequencing gaps. For Nanopore, low coverage regions with poor alignment quality (<85% variant homogeneity) near sequencing/amplicon ends were masked to be robust against primer drop-out experienced in the Spike gene, and the sensitivity for detecting short inserts using a region-local global alignment of reads, was increased. In addition, we also used the wf_artic (ARTIC SARS-CoV-2) pipeline as built using the nextflow workflow framework57. In some instances, mutations were confirmed visually with .bam files using Geneious software V2020.1.2 (Biomatters). The reference genome used throughout the assembly process was NC_045512.2 (numbering equivalent to MN908947.3). For lineage classification, we used the widespread dynamic lineage classification method from the 'Phylogenetic Assignment of Named Global Outbreak Lineages' (PANGOLIN) software suite (https://github.com/hCoV-2019/pangolin).

Cells

Vero E6 cells (originally ATCC CRL-1586, obtained from Cellonex in South Africa) were propagated in complete growth medium consisting of Dulbecco's Modified Eagle Medium (DMEM) with 10% fetal bovine serum (Hyclone) containing 10mM of HEPES, 1mM sodium pyruvate, 2mM L-glutamine and 0.1mM nonessential amino acids (Sigma-Aldrich). Vero E6 cells were passaged every 3–4 days. The H1299-E3 cell line (H1299 originally from ATCC as CRL-5803) was propagated in growth medium consisting of complete Roswell Park Memorial Institute (RPMI) 1640 medium with 10% fetal bovine serum containing 10mM of HEPES, 1mM sodium pyruvate, 2mM L-glutamine and 0.1mM nonessential amino acids. Cells were passaged every second day. The H1299-E3 (H1299-ACE2, clone E3) cell line was derived from H1299 as described in our previous work[1,17].

Virus expansion

All work with live virus was performed in Biosafety Level 3 containment using protocols for SARS-CoV-2 approved by the Africa Health Research Institute Biosafety Committee. ACE2-expressing H1299-E3

cells were seeded at 4.5×10^5 cells in a 6 well plate well and incubated for 18–20 h. After one DPBS wash, the sub-confluent cell monolayer was inoculated with 500 µL universal transport medium diluted 1:1 with growth medium filtered through a 0.45-µm filter. Cells were incubated for 1 h. Wells were then filled with 3 mL complete growth medium. After 4 days of infection (completion of passage 1 (P1)), cells were trypsinized, centrifuged at 300 rcf for 3 min and resuspended in 4 mL growth medium. Then all infected cells were added to Vero E6 cells that had been seeded at 2×10^5 cells per mL, 20mL total, 18–20 h earlier in a T75 flask for cell-to-cell infection. The coculture of ACE2-expressing H1299-E3 and Vero E6 cells was incubated for 1 h and the flask was filled with 20 mL of complete growth medium and incubated for 4 days. The viral supernatant from this culture (passage 2 (P2) stock) was used for experiments.

Live virus neutralization assay

H1299-E3 cells were plated in a 96-well plate (Corning) at 30,000 cells per well 1 day pre-infection. Plasma was separated from EDTA-anticoagulated blood by centrifugation at 500 rcf for 10 min and stored at −80 °C. Aliquots of plasma samples were heat-inactivated at 56 °C for 30 min and clarified by centrifugation at 10,000 rcf for 5 min. Virus stocks were used at approximately 50-100 focus-forming units per microwell and added to diluted plasma. Antibody–virus mixtures were incubated for 1 h at 37 °C, 5% CO_2. Cells were infected with 100 µL of the virus–antibody mixtures for 1 h, then 100 µL of a 1X RPMI 1640 (Sigma-Aldrich, R6504), 1.5% carboxymethylcellulose (Sigma-Aldrich, C4888) overlay was added without removing the inoculum. Cells were fixed 18 h post-infection using 4% PFA (Sigma-Aldrich) for 20 min. Foci were stained with a rabbit anti-spike monoclonal antibody (BS-R2B12, GenScript A02058) at 0.5 µg/mL in a permeabilization buffer containing 0.1% saponin (Sigma-Aldrich), 0.1% BSA (Sigma-Aldrich) and 0.05% Tween-20 (Sigma-Aldrich) in PBS. Plates were incubated with primary antibody overnight at 4 °C, then washed with wash buffer containing 0.05% Tween-20 in PBS. Secondary goat anti-rabbit HRP conjugated antibody (Abcam ab205718) was added at 1 µg/mL and incubated for 2 h at room temperature with shaking. TrueBlue peroxidase substrate (SeraCare 5510-0030) was then added at 50 µL per well and incubated for 20 min at room temperature. Plates were imaged in an ImmunoSpot Ultra-V S6-02-6140 Analyzer ELISPOT instrument with BioSpot Professional built-in image analysis (C.T.L).

Statistics and fitting

All statistics and fitting were performed using custom code in MATLAB v.2019b. Neutralization data were fit to:

$$Tx=1/1+(D/ID_{50}).$$

Here Tx is the number of foci normalized to the number of foci in the absence of plasma on the same plate at dilution D and ID_{50} is the plasma dilution giving 50% neutralization. $FRNT_{50} = 1/ID_{50}$. Values of $FRNT_{50} < 1$ are set to 1 (undiluted), the lowest measurable value. We note that the most concentrated plasma dilution was 1:25 and therefore $FRNT_{50} < 25$ were extrapolated. To calculate confidence intervals, $FRNT_{50}$ or fold-change in $FRNT_{50}$ per participant was log transformed and arithmetic mean plus 2 std and arithmetic mean minus 2 std were calculated for the log transformed values. These were exponentiated to obtain the upper and lower 95% confidence intervals on the geometric mean $FRNT_{50}$ or the fold-change in $FRNT_{50}$ geometric means.

Acknowledgements

This study was supported by the Bill and Melinda Gates award INV-018944 (AS), National Institutes of Health award R01 AI138546 (AS), and South African Medical Research Council awards including 6084COAP2020 (AS, TdO, PLM) and the UK Foreign, Commonwealth and Development Office and Wellcome Trust (Grant no 221003/Z/20/Z, PLM). PLM is also supported by the South African Research Chairs Initiative of the Department of Science and Innovation and the NRF (Grant No 98341). The

funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Materials and methods references

29 Cele, S. et al. Omicron extensively but incompletely escapes Pfizer BNT162b2 neutralization. Nature, doi:10.1038/s41586-021-04387-1 (2021).

30 Cele, S. et al. SARS-CoV-2 prolonged infection during advanced HIV disease evolves extensive immune escape. Cell Host & Microbe, doi:10.1016/j.chom.2022.01.005 (2022).

31 Cele, S. et al. Escape of SARS-CoV-2 501Y.V2 from neutralization by convalescent plasma. Nature 593, 142-146, doi:10.1038/s41586-021-03471-w (2021).

Figure Legends

Figure 1: Enhancement of Delta neutralization by Omicron infection. Neutralization of Omicron/BA.1 virus was determined for (a) n=15 Omicron/BA.1 infected vaccinated or (b) n=24 Omicron/BA.1 infected unvaccinated participants. Neutralization of Delta virus was determined for the same (c) vaccinated (n=15) and (d) unvaccinated (n=24) participants. Each participant was sampled at the initial enrollment visit (median 6 days post-symptom onset) and compared to the last follow-up visit (median 23 days post-symptom onset). Neutralization capacity per participant was determined in two independent experiments and numbers are geometric mean titers (GMT) over all participants per group of the reciprocal plasma dilution ($FRNT_{50}$) resulting in 50% neutralization, Fold-change is calculated by dividing the follow-up by the enrollment visit GMT. Dashed line is most concentrated plasma tested. p-values were determined by a left-sided Wilcoxon rank sum test measuring significance of increase and represented as **0.01-0.001 or ns, not significant. Exact p-values were (a) 0.0012, (b) 0.0081, (c) 0.0021, (d) 0.11.

Figure 2: Gap in neutralizing immunity between Omicron/BA.1 infected vaccinated and unvaccinated participants. (a) Neutralization of Omicron/BA.2, Beta, Delta, and ancestral virus (with the D614G substitution) compared to Omicron/BA.1 virus at the last available follow-up timepoint in n=15 Omicron/BA.1 infected vaccinated or n=24 Omicron/BA.1 infected unvaccinated participants. Neutralization capacity per participant was determined in two independent experiments and numbers are GMT $FRNT_{50}$. Fold-change was calculated by dividing the larger by the smaller GMT. Dashed line is most concentrated plasma tested. p-values were determined by a two-sided Wilcoxon rank sum test and represented as *0.05-0.01, ns, not significant. Exact p-values (vaccinated/unvaccinated) were: 0.22/0.087 for BA.2, 0.36/0.071 for Beta, 0.15/0.25 for Delta, and 0.014/0.20 for ancestral. (b) Comparison of neutralization capacity in the Omicron/BA.1 infected vaccinated (n=15) versus Omicron/BA.1 infected unvaccinated (n=24) participants against Omicron/BA.1, Omicron/BA.2, Beta, Delta and ancestral/D614G viruses. Neutralization capacity per participant was determined in two independent experiments for all strains except for Omicron/BA.1, where 6 experiments were available and were used in the calculation. Points represent GMT $FRNT_{50}$ per group and error bars are GMT 95% confidence intervals. p-values were determined by a two-sided Wilcoxon rank sum test and represented as *0.05-0.01, **0.01-0.001, ***0.001-0.0001. Exact p-values were 0.025 (BA.1), 0.0026 (BA.2), 4.1 X 10^{-4} (Beta), 0.0012 (Delta), 3.3 X 10^{-4} (ancestral).

Figure 3: Escape of Omicron virus from Delta infection elicited immunity. Neutralization of Delta compared to Omicron/BA.1 virus by Delta infection elicited plasma immunity in vaccinated and

unvaccinated participants. 18 samples were tested from n=14 participants infected during the Delta infection wave in South Africa. Dashed line is the most concentrated plasma tested. p-value (****) is 3.2×10^{-7} as determined by a two-sided Wilcoxon rank sum test.

Extended Data Table 1: Characteristics of Omicron/BA.1 infected participants

Ct enrol.: qPCR cycle threshold for SARS-CoV-2 at enrollment. Symptoms to enrol.: time between symptoms onset and study enrolment. Symp. to last follow-up: time between symptoms onset and last follow-up visit. Max CD4: maximum CD4 count per microliter blood across all study visits. Supp O_2: participant required supplemental oxygen during the study. Hosp.: participant hospitalized during the study. UND: Undetectable Ct. N/A: Not available; sequencing failed, usually due to insufficient virus substrate. *Reported previous infection. **Boosted with Ad26.CoV2.S in Nov-2021. ***Participants with persistent low CD4 count and uncontrolled HIV viremia indicative of advanced HIV disease and immune suppression. Excluded from analysis. [$]Deceased.

Extended Data Table 2: Summary characteristics of Omicron/BA.1 infected participants

Values are median (IQR). Hospital stay calculated to last inpatient study visit.

Extended Data Table 3: Characteristics of Delta infected participants

*Asymptomatic, date of diagnostic swab used instead of symptoms onset. Ct enrol: qPCR cycle threshold for SARS-CoV-2 at enrollment. UND: undetectable. Pre: sample taken pre-vaccination. Post: sample taken post-vaccination for participants with a pre-vaccination sample. N/A: not available.

Extended Data Figure 1: Longitudinal Omicron/BA.1 and Delta neutralization capacity in Omicron/BA.1 infected participants. Neutralization of Omicron (blue) and Delta (red) at all study visits. Participant number is as in Extended Data Table 1. Top three rows are participants vaccinated with Pfizer BNT162b2 (n=8) or Johnson and Johnson Ad26.CoV2.S (n=7) and bottom five rows are unvaccinated participants (n=24). X-axis is the time post-symptom onset when sample was collected, and y-axis is neutralization as $FRNT_{50}$. Dashed line is the most concentrated plasma tested (LOQ, limit of quantification below which $FRNT_{50}$ values are extrapolated). All participants recovered except participant 29, who died.

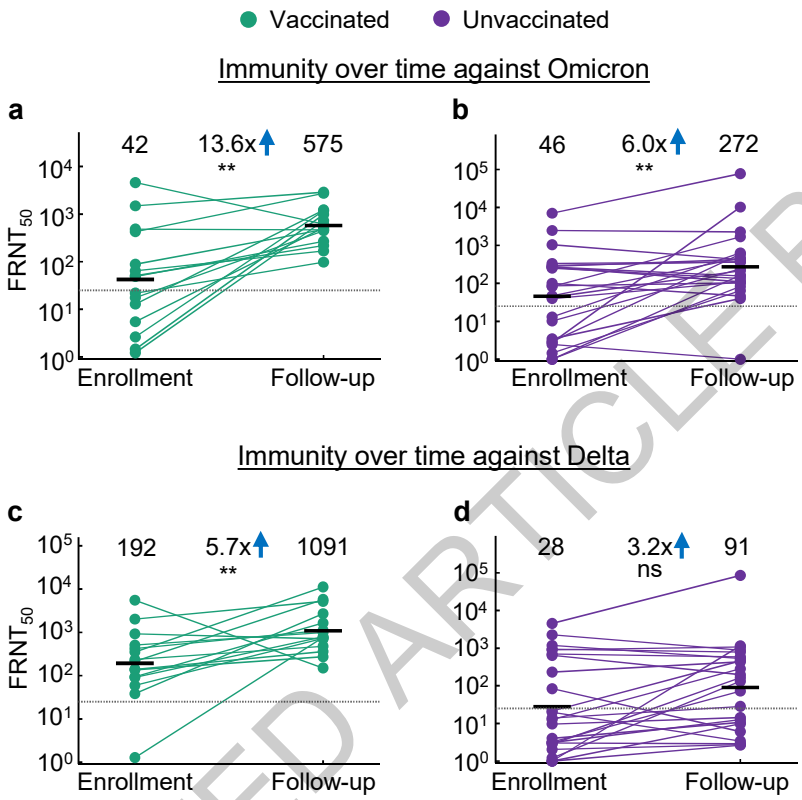
Extended Data Figure 2: Fold-drop in BA.2 versus BA.1 neutralization in all and sequence confirmed samples. Neutralization of Omicron BA.2 compared to BA.1 in participants described in Extended Data Table 1, excluding participant 14 for technical reasons and participants 40 and 41 because of advanced HIV disease. Left panel shows neutralization capacity in all n=38 participants and right panel shows neutralization capacity for n=25 participants where infection was successfully sequenced and determined to be BA.1. Dashed line is the most concentrated plasma tested. p-values were 0.077 for all and 0.15 for BA.1 sequence confirmed participants as determined by a two-sided Wilcoxon rank sum test. ns, not significant.

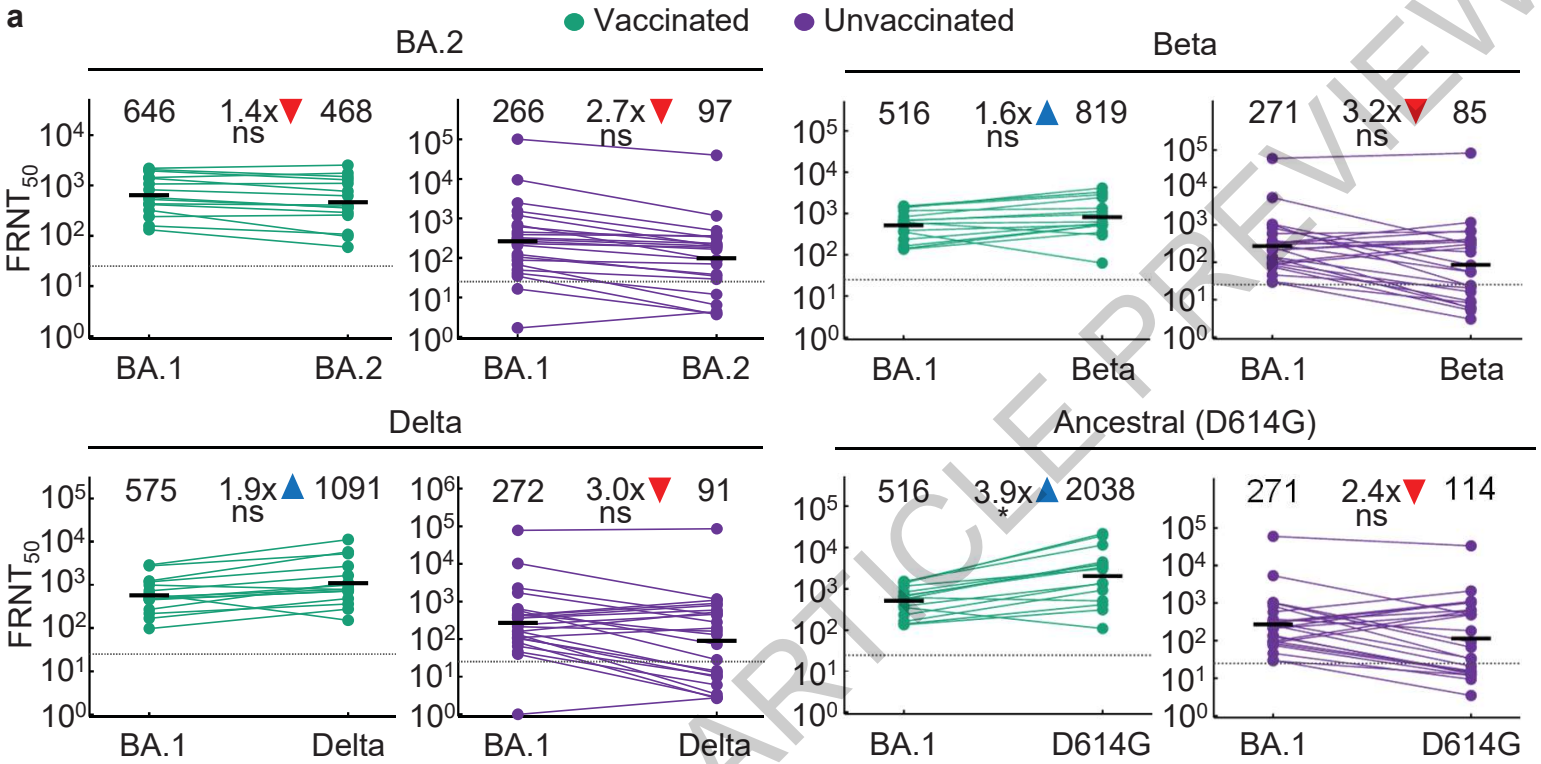
COMMIT-KZN Team

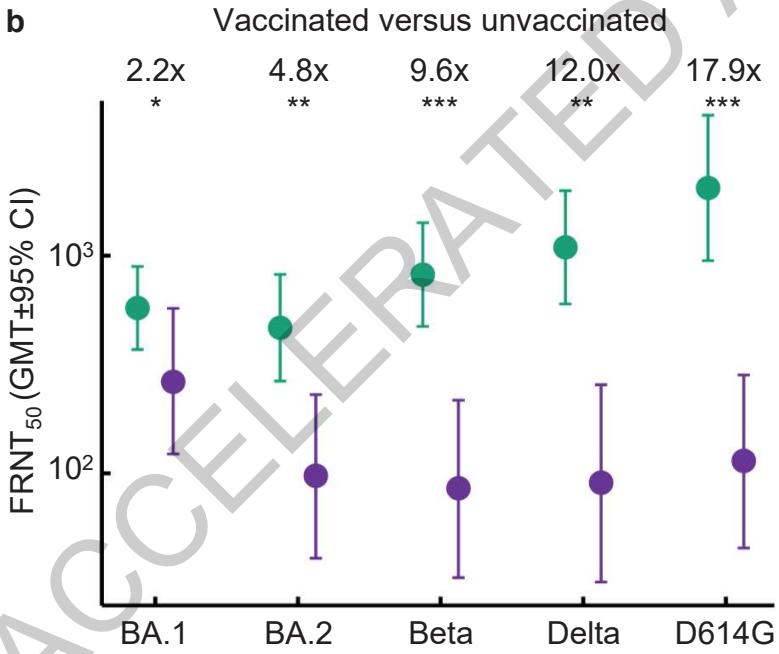
Rohen Harrichandparsad[18], Kobus Herbst[1,19], Prakash Jeena[20], Thandeka Khoza[1], Henrik Kløverpris[1,21], Alasdair Leslie[1,13], Rajhmun Madansein[22], Mohlopheni Marakalala[1,13], Mosa Moshabela[23], Kogie Naidoo[4], Zaza Ndhlovu[1, 24], Thumbi Ndung'u[1,17,24,25], Kennedy Nyamande[26], Vinod Patel[27], Theresa Smit[1], Adrie Steyn[1,28] & Emily Wong[1,28].

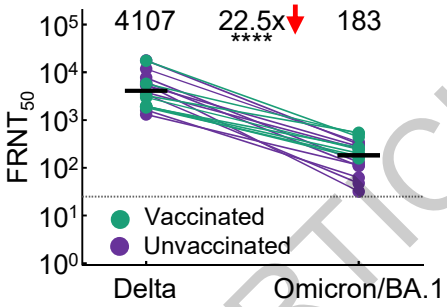
[18]Department of Neurosurgery, University of KwaZulu-Natal, Durban, South Africa. [19]South African Population Research Infrastructure Network, Durban, South Africa. [20]Department of Paediatrics and Child Health, University of KwaZulu-Natal, Durban, South Africa. [21]Department of Immunology and Microbiology, University of Copenhagen, Copenhagen, Denmark. [22]Department of Cardiothoracic Surgery, University of KwaZulu-Natal, Durban, South Africa.[23]College of Health Sciences, University of KwaZulu-Natal, Durban, South Africa. [24]Ragon Institute of MGH, MIT and Harvard, Boston, USA. [25]HIV Pathogenesis Programme, The Doris Duke Medical Research Institute, University of KwaZulu-

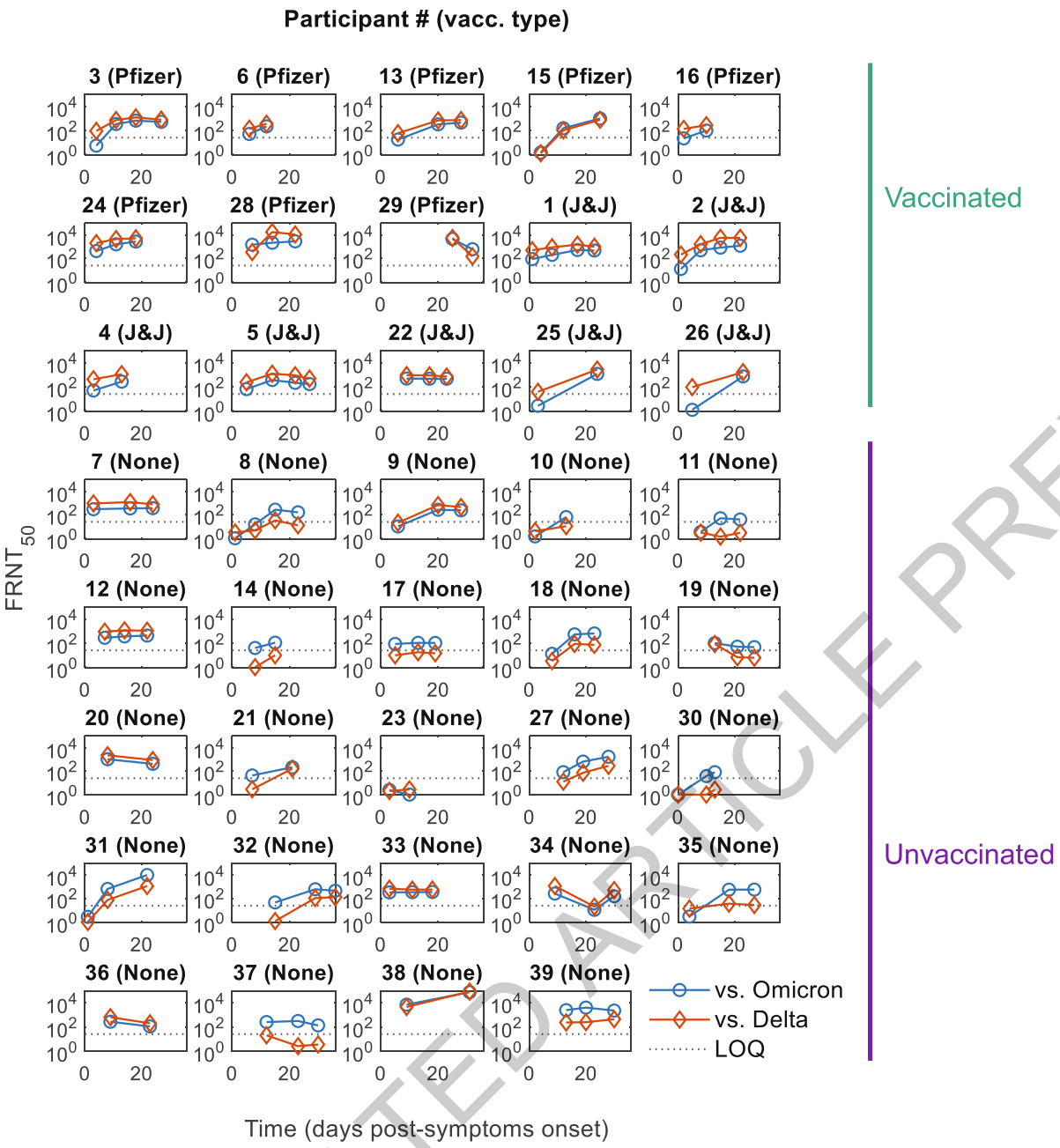
464 Natal, Durban, South Africa. [26]Department of Pulmonology and Critical Care, University of KwaZulu-
465 Natal, Durban, South Africa. [27]Department of Neurology, University of KwaZulu-Natal, Durban, South
466 Africa.[28]Division of Infectious Diseases, University of Alabama at Birmingham, USA



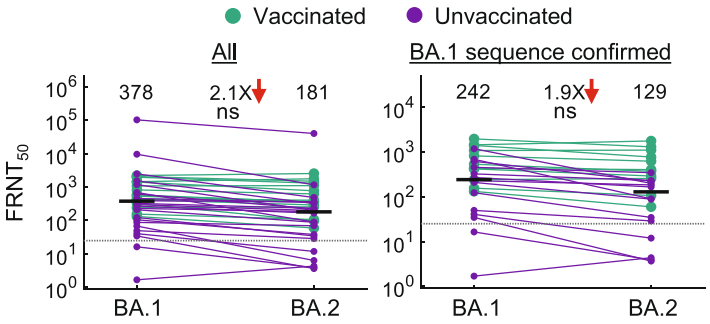








Extended Data Fig. 1



Extended Data Fig. 2

#	Age	Sex	Vacc. type	Date of vacc.	Vacc. to enrol. (days)	Date symp. onset	Ct at enrol.	Symp. to enrol. (days)	Symp. to last follow-up (days)	Max CD4	Sub-lineage	Seq. GISAID ID	Supp. O_2	Hosp.
1	30-39	M	AD26.COV2	MAR-2021	274	DEC-2021*	24.9	1	23	1071	BA.1	EPI_ISL_9967759	No	No
2	30-39	M	AD26.COV2*	NOV-2021	14	NOV-2021	14.5	1	22	789	BA.1	EPI_ISL_9967761	No	No
3	50-59	F	BNT162b2	JUL-2021	138	DEC-2021	16.8	4	27	777	BA.1	EPI_ISL_8604915	No	No
4	30-39	F	AD26.COV2	MAY-2021	210	DEC-2021	30.7	3	13	1169	BA.1	EPI_ISL_8604910	No	No
5	20-29	F	AD26.COV2	SEP-2021	89	DEC-2021	23.9	5	27	1220	BA.1	EPI_ISL_9967760	No	Yes
6	10-19	F	BNT162b2	JUL-2021	157	DEC-2021	23.1	6	12	732	BA.1	EPI_ISL_8604906	No	Yes
7	20-29	F				NOV-2021	UND	3	24	712	N/A	N/A	No	Yes
8	30-39	M				DEC-2021	18.2	1	23	847	BA.1	EPI_ISL_8604919	No	Yes
9	40-49	F				DEC-2021	32.3	6	28	1032	BA.1	EPI_ISL_8604901	No	Yes
10	20-29	M				DEC-2021	30.4	2	13	1197	BA.1	EPI_ISL_8604908	No	Yes
11	20-29	F				DEC-2021	28.3	8	22	863	BA.1	EPI_ISL_8604913	No	No
12	20-29	F				DEC-2021*	UND	7	22	1259	BA.1	EPI_ISL_8604912	No	Yes
13	30-39	M	BNT162b2	JUL-2021	129	NOV-2021	31.6	6	28	1069	BA.1	EPI_ISL_8604916	No	Yes
14	20-29	M				NOV-2021	30.8	8	15	1225	N/A	N/A	No	Yes
15	60-69	F	BNT162b2	JUL-2021	139	DEC-2021	24.6	4	25	345	BA.1	EPI_ISL_8604920	No	Yes
16	60-69	M	BNT162b2	DEC-2021	15	DEC-2021	24.6	2	10	904	BA.1	EPI_ISL_8578311	No	No
17	30-39	M				DEC-2021	37.0	5	19	1008	BA.1	EPI_ISL_8604923	No	No
18	60-69	F				DEC-2021	26.8	8	23	1111	BA.1	EPI_ISL_8578312	Yes	Yes
19	30-39	M				DEC-2021*	30.7	13	27	1077	BA.1	EPI_ISL_8604924	No	Yes
20	20-29	F				DEC-2021	35.9	8	24	533	BA.1	EPI_ISL_8604911	No	Yes
21	20-29	M				DEC-2021	29.1	7	21	225	BA.1	EPI_ISL_8604922	No	No
22	30-39	F	AD26.COV2	AUG-2021	120	DEC-2021	33.4	9	23	777	BA.1	EPI_ISL_8693907	No	Yes
23	20-29	F				DEC-2021	35.8	3	10	1167	BA.1	EPI_ISL_8604902	No	No
24	50-59	M	BNT162b2	AUG-2021	128	DEC-2021	36.6	4	18	605	N/A	N/A	No	Yes
25	30-39	F	AD26.COV2	APR-2021	237	DEC-2021	23.5	3	24	640	BA.1	EPI_ISL_8604914	No	No
26	50-59	F	AD26.COV2	JUL-2021	150	DEC-2021	UND	5	23	716	N/A	N/A	No	No
27	50-59	F				DEC-2021	32.4	12	28	625	N/A	N/A	Yes	Yes
28	80-89	F	BNT162b2	JUL-2021	177	JAN-2022	30.8	7	22	407	N/A	N/A	Yes	Yes
29	60-69	M	BNT162b2	JUL-2021	178	DEC-2021[$]	UND	25	32	351	N/A	N/A	Yes	Yes
30	40-49	M				DEC-2021	20.2	0	13	844	BA.1	EPI_ISL_8604909	No	No
31	30-39	F				DEC-2021	34.8	1	22	647	N/A	N/A	Yes	Yes
32	50-59	F				DEC-2021	28.2	15	36	620	BA.1	EPI_ISL_8578347	No	Yes
33	20-29	F				DEC-2021	UND	3	18	902	N/A	N/A	No	Yes
34	30-39	F				DEC-2021	34.8	9	30	1363	N/A	N/A	No	Yes
35	50-59	F				DEC-2021	26.6	4	27	766	BA.1	EPI_ISL_8578342	Yes	Yes
36	20-29	F				DEC-2021	UND	9	23	1212	N/A	N/A	No	Yes
37	50-59	F				DEC-2021	UND	12	30	995	N/A	N/A	No	Yes
38	30-39	M				DEC-2021	UND	9	31	746	N/A	N/A	No	Yes
39	50-59	F				DEC-2021	UND	13	30	840	N/A	N/A	Yes	Yes
40	30-39	F				DEC-2021	22.5	5	19	61***	BA.1	EPI_ISL_8578314	Yes	Yes
41	40-49	F				NOV-2021	29.8	17	24	53***	N/A	N/A	No	Yes

Extended Data Table 1

	All 39	Vaccinated 15 (38%)	Unvaccinated 24 (62%)
Age	35 (27-55)	37 (32-60)	31.5 (26-49)
Female	25 (64%)	9 (60%)	16 (67%)
Vaccination to enrollment (days)	-	139 (120-178)	-
Symptom onset to enrolment (days)	6 (3-9)	4 (3-6)	7.5 (3-9)
Symptom onset to last follow-up (days)	23 (19-27)	23 (18-27)	23 (20-28)
Maximum CD4 count (cell/μL)	844 (647-1077)	777 (605-1069)	882.5 (729-1139)
Required supp. O_2	7 (18%)	2 (13%)	5 (21%)
Hospitalized	27 (69%)	8 (53%)	19 (79%)
Duration of hospitalization (days)	7 (3-11)	3.5 (2.5-14.5)	8 (3-11)

Extended Data Table 2

#	Age	Sex	Vacc. type	Date of vacc.	Vacc. to enrol. (days)	Date symp. onset	Ct at enrol.	Symp. to collection (days)	Seq. GISAID ID
1	40-49	F				JUL-2021	26	26	EPI_ISL_3722338
2	40-49	M				JUL-2021	31	23^	EPI_ISL_3722335
3	50-59	M				JUL-2021	30	31	N/A
4	50-59	M				JUN-2021	27	37	N/A
5	40-49	M				JUL-2021	35	44	N/A
6	30-39	M				JUL-2021	37	32	N/A
7	70-79	M	BNT162b2	JUN-2021	37	JUL-2021	37	15	N/A
8	60-69	F	BNT162b2	NOV-2021	14	AUG-2021	UND	116	N/A
9	40-49	F	AD26.COV	MAY-2021	117	JUL-2021	UND	31	N/A
10	50-59	F	AD26.COV	APR-2021	147	JUL-2021	UND	57	N/A
11 Pre	40-49	M				AUG-2021*	35	13*	N/A
11 Post	40-49	M	BNT162b2	OCT-2021	18	AUG-2021	UND	83	N/A
12 Pre	40-49	M				JUL-2021	23	24	EPI_ISL_3939068
12 Post	40-49	M	AD26.COV	SEP-2021	32	JUL-2021	UND	92	N/A
13 Pre	30-39	M				JUL-2021	27	24	EPI_ISL_3939088
13 Post	30-39	M	AD26.COV	SEP-2021	32	JUL-2021	UND	94	N/A
14 Pre	50-59	F				JUL-2021*	27	23*	EPI_ISL_3447779
14 Post	50-59	F	BNT162b2	OCT-2021	22	JUL-2021	UND	93	N/A

Extended Data Table 3

nature portfolio

Corresponding author(s): Alex Sigal

Last updated by author(s): Apr 18, 2022

Reporting Summary

Nature Portfolio wishes to improve the reproducibility of the work that we publish. This form provides structure for consistency and transparency in reporting. For further information on Nature Portfolio policies, see our Editorial Policies and the Editorial Policy Checklist.

Statistics

For all statistical analyses, confirm that the following items are present in the figure legend, table legend, main text, or Methods section.

n/a	Confirmed	
☐	☒	The exact sample size (*n*) for each experimental group/condition, given as a discrete number and unit of measurement
☐	☒	A statement on whether measurements were taken from distinct samples or whether the same sample was measured repeatedly
☐	☒	The statistical test(s) used AND whether they are one- or two-sided *Only common tests should be described solely by name; describe more complex techniques in the Methods section.*
☒	☐	A description of all covariates tested
☒	☐	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons
☐	☒	A full description of the statistical parameters including central tendency (e.g. means) or other basic estimates (e.g. regression coefficient) AND variation (e.g. standard deviation) or associated estimates of uncertainty (e.g. confidence intervals)
☐	☒	For null hypothesis testing, the test statistic (e.g. *F*, *t*, *r*) with confidence intervals, effect sizes, degrees of freedom and *P* value noted *Give P values as exact values whenever suitable.*
☒	☐	For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings
☒	☐	For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes
☒	☐	Estimates of effect sizes (e.g. Cohen's *d*, Pearson's *r*), indicating how they were calculated

Our web collection on statistics for biologists contains articles on many of the points above.

Software and code

Policy information about availability of computer code

Data collection	Participant data was collected using REDCap version 11.1.29
Data analysis	All statistics and fitting were performed using MATLAB v.2019b

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors and reviewers. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Portfolio guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a data availability statement. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A description of any restrictions on data availability
- For clinical datasets or third party data, please ensure that the statement adheres to our policy

Sequences of outgrown Omicron sub-lineages have been deposited in GISAID with accession EPIISL_7886688 (Omicron/BA.1) and EPIISL9082893 (Omicron/BA.2). Delta, Beta, and ancestral SARS-CoV-2 isolates have been previously described (15). Raw images of the data are available upon reasonable request.

Field-specific reporting

Please select the one below that is the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

☒ Life sciences ☐ Behavioural & social sciences ☐ Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/documents/nr-reporting-summary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Samples size was not predetermined. We offered study enrollment to all participants who met the inclusion/exclusion criteria
Data exclusions	We excluded two samples from participants who were immune suppressed due to advanced HIV disease based on a low CD4 count and uncontrolled HIV viremia.
Replication	Repeated in independent experiments on different days in sets of paired experiments to which always included Omicron/BA.1. Two experiments performed for all variants and 6 experiments performed for BA.1. Geometric mean of measurements per participant was used.
Randomization	No randomization.
Blinding	No blinding

Reporting for specific materials, systems and methods

We require information from authors about some types of materials, experimental systems and methods used in many studies. Here, indicate whether each material, system or method listed is relevant to your study. If you are not sure if a list item applies to your research, read the appropriate section before selecting a response.

Materials & experimental systems

n/a	Involved in the study
☐	☒ Antibodies
☐	☒ Eukaryotic cell lines
☒	☐ Palaeontology and archaeology
☒	☐ Animals and other organisms
☐	☒ Human research participants
☒	☐ Clinical data
☒	☐ Dual use research of concern

Methods

n/a	Involved in the study
☒	☐ ChIP-seq
☒	☐ Flow cytometry
☒	☐ MRI-based neuroimaging

Antibodies

Antibodies used	Foci were stained with a rabbit anti-spike monoclonal antibody (BS-R2B12, GenScript A02058) at 0.5 µg/mL. Secondary goat anti-rabbit horseradish peroxidase (Abcam ab205718) antibody was added at 1 µg/mL
Validation	Information sheet for A02058 at https://www.genscript.com/antibody/A02058-MonoRab_SARS_CoV_2_Spike_S1_Antibody_BS_R2B12_mAb_Rabbit.html. Information sheet for ab205718: https://www.abcam.com/goat-rabbit-igg-hl-hrp-ab205718.html

Eukaryotic cell lines

Policy information about cell lines

Cell line source(s)	Vero E6 cells (ATCC CRL-1586) obtained from Cellonex in South Africa. The H1299-E3 (H1299-ACE2, clone E3) cell line was derived from H1299 as described in our previous work. H1299 was a gift from M. Oren, originally obtained from ATCC (CRL-5803)
Authentication	Cell lines have not been authenticated.
Mycoplasma contamination	The cell lines have been tested for mycoplasma contamination and are mycoplasma negative.
Commonly misidentified lines (See ICLAC register)	None

Human research participants

Policy information about studies involving human research participants

Population characteristics	Participant characteristics are listed per participant in Table S1 and S3 and summarized in Table S2.
Recruitment	Blood samples were obtained from hospitalized adults with PCR-confirmed SARS-CoV-2 infection and/or vaccinated individuals who were enrolled in a prospective cohort study approved by the Biomedical Research Ethics Committee at the University of KwaZulu–Natal. Investigators were blinded to participant information.
Ethics oversight	Study approved by the Biomedical Research Ethics Committee at the University of KwaZulu–Natal (reference BREC/00001275/2020). Use of residual Omicron/BA.1 swab sample was approved by the University of the Witwatersrand Human Research Ethics Committee (HREC) (ref. M210752). Use of swab sample to isolate Omicron/BA.2 was collected as part of the "COVID-19 transmission and natural history in KwaZulu-Natal, South Africa: Epidemiological Investigation to Guide Prevention and Clinical Care" Centre for the AIDS Programme of Research in South Africa (CAPRISA) study and approved by the Biomedical Research Ethics Committee at the University of KwaZulu–Natal (reference 201 BREC/00001195/2020, BREC/00003106/2021).

Note that full information on the approval of the study protocol must also be provided in the manuscript.