HUMAN VACCINES & IMMUNOTHERAPEUTICS
2024, VOL. 20, NO. 1, 2423466
https://doi.org/10.1080/21645515.2024.2423466

Taylor & Francis
Taylor & Francis Group

RESEARCH ARTICLE

OPEN ACCESS Check for updates

Antibody dynamics for heterologous boosters with aerosolized Ad5-nCoV following inactivated COVID-19 vaccines

Canjie Chen[a*], Tian Tang[a*], Zhao Chen[a*], Lan Chen[a,b], Jinling Cheng[a], Fang Li[a], Jing Sun[a], Jingxian Zhao[a,b], Yanqun Wang[a], Qihong Yan[a], Jincun Zhao[a,b,c,d], and Airu Zhu[a]

[a]State Key Laboratory of Respiratory Disease, National Clinical Research Centre for Respiratory Disease, Guangzhou Institute of Respiratory Health, The First Affiliated Hospital of Guangzhou Medical University, Guangzhou, China; [b]Department of Basic Research, Guangzhou National Laboratory, Guangzhou, China; [c]Shanghai Institute for Advanced Immunochemical Studies, School of Life Science and Technology, Shang-haiTech University, Shanghai, China; [d]National Clinical Research Center for Infectious Disease, Shenzhen Third People's Hospital, and The Second Affiliated Hospital, School of Medicine, Southern University of Science and Technology, Shenzhen, China

ABSTRACT

The COVID-19 pandemic has underscored vaccination as a crucial strategy for reducing disease severity and preventing hospitalizations. Heterologous boosters using aerosolized Ad5-nCoV following two doses of inactivated vaccine have demonstrated superior antibody responses. However, the comprehensive dynamics of this antibody boost and the optimal timing for heterologous boosters are still not fully understood. In this study, we investigated the dynamics of neutralizing antibody (nAb) responses in recipients of heterologous booster vaccinations with aerosolized Ad5-nCoV following either two (I-I-A) or three (I-I-I-A) doses of COVID-19 inactivated vaccines. The findings indicate that a booster dose of aerosolized Ad5-nCoV vaccine induced robust and durable nAb responses comparable to those elicited in BA.5 breakthrough infections with similar doses of inactivated vaccine. Notably, group I-I-A showed higher peak nAb titers against the WT strain, BA.5, and XBB.1 variants compared to group I-I-I-A, inversely correlating with the prior nAb levels. This suggesting the possible efficacy of the heterologous aerosolized Ad5-nCoV booster and indicates that pre-boost antibody levels may be related to the outcomes of booster vaccination.

ARTICLE HISTORY
Received 26 July 2024
Revised 21 October 2024
Accepted 26 October 2024

KEYWORDS
COVID-19; inhaled adenovirus type 5 vector vaccines; vaccination strategies; antibodies; vaccine response efficacy

Introduction

The COVID-19 pandemic has highlighted vaccination as a pivotal intervention for reducing disease severity and preventing hospitalizations.[1,2] Since 2020, numerous vaccines have been developed and deployed worldwide to combat the pandemic. As of March 30, 2023, the World Health Organization (WHO) reported 183 COVID-19 vaccines in clinical development and 199 in pre-clinical development. These vaccines are based on various platforms, including inactivated virus, mRNA, protein subunit, and viral vectored vaccines.[3] Most approved vaccines are administered via intramuscular injections, which primarily induce serological IgG. However, intramuscular vaccination does not provide a robust first line of defense in the respiratory tract due to a lack of mucosal immunity. This limitation underscores the need for inhalable or intranasal vaccines that mimic the natural entry route of SARS-CoV-2 via the airways.[4–7]

To address this issue, several viral vectored vaccines, including those using adenoviruses[8] or attenuated influenza viruses[9] for intranasal or inhaled administration, have been approved. Remarkably, clinical studies have demonstrated that inhaled vaccines utilizing the adenovirus type 5 vector (Ad5) can generate antibody responses comparable to those induced by intramuscular injections, but at significantly lower doses.[8] Moreover, the inhaled Ad5 vector COVID-19 vaccine (Ad5-nCoV) has been reported to be safe and capable of inducing higher levels of neutralizing antibodies (nAbs) against the prototype and Omicron BA.5 variant following a sequential vaccination strategy after two doses of inactivated vaccines (I-I-A).[10–12] However, the effectiveness of the aerosol Ad5-nCoV as a sequential booster after three doses of inactivated vaccines remains unknown. Additionally, the optimal sequential strategy is still under investigation.

In this study, we describe the kinetics of antibody immune responses induced by two different vaccination strategies: a heterologous booster with aerosol Ad5-nCoV vaccine after two (I-I-A) or three (I-I-I-A) doses of inactivated vaccines. We compared the neutralizing antibody responses elicited by the heterologous aerosolized Ad5-nCoV booster to those observed in BA.5 breakthrough infections. Furthermore, we explored the factors contributing to the differences in nAb responses between the I-I-A and I-I-I-A groups. Our findings provide valuable insights for selecting effective vaccination strategies and suggest that pre-boost antibody levels could be used to predict the efficacy of booster vaccination.

CONTACT Qihong Yan yan_qihong@126.com; Jincun Zhao zhaojincun@gird.cn; Airu Zhu zhuairu14@163.com State Key Laboratory of Respiratory Disease, National Clinical Research Centre for Respiratory Disease, Guangzhou Institute of Respiratory Health, the First Affiliated Hospital of Guangzhou Medical University, 195 Dongfeng Xi Road, Yuexiu District, Guangzhou 510182, Guangdong, China
*Co-first authors.

Supplemental data for this article can be accessed on the publisher's website at https://doi.org/10.1080/21645515.2024.2423466

Materials and methods

Study cohort

The inhaled aerosol Ad5-nCoV vaccines contain a replication-defective adenovirus type 5 (Ad5) vector expressing the full-length spike protein gene of the wild-type SARS-CoV-2, strain Wuhan-Hu-1. To assess efficiency of heterologous booster vaccination following two or three doses of inactivated vaccines, we recruited 35 participants who inhaled an aerosol Ad5-nCoV as booster vaccination, consist of 18 who had finished two doses of inactivated vaccines (I-I-A group; 12 [66.7%] female, 6 [33.3%] male; median age 27 years [22 ~ 49]), and 17 with three doses (I-I-I-A group; 13 [76.5%] female, 4 [23.5%] male; median age 26 years [22 ~ 35]). Additionally, we enrolled 112 patients who had experienced BA.5 breakthrough infections during August 2022 to January 2023, including 33 had received two doses of an inactivated vaccine before the infection (I-I-Bi group; 23 [69.7%] female, 10 [30.3%] male; median age 38 years [4 ~ 89]), and 79 had received three doses of an inactivated vaccine before the infection (I-I-I-Bi group; 37 [46.8%] female, 42 [53.2%] male; median age 51 years [20 ~ 84]; Table 1). Plasma samples were collected from each subject at multiple time points. (Figure 1a).

Focus reduction neutralization test

The SARS-CoV-2 focus reduction neutralization test (FRNT) was performed in a certified Biosafety Level 3 (BSL-3) laboratory as previously described.[13] Plasma samples (50 µL) were serially diluted and combined with an equal volume of SARS-CoV-2 virus (100 focus forming units, FFU) in 96-well plates, followed by a 1-hour incubation at 37°C. The mixtures were then added to Vero E6 cell-seeded 96-well plates (ATCC, Manassas, VA) and incubated for an additional hour at 37°C to facilitate virus entry. After removing the inoculum, overlay media (100 µL MEM with 1.2% carboxymethyl cellulose, CMC) was added, and the plates were incubated at 37°C for 24 hours. Following incubation, the overlays were removed, and cells were fixed with 4% paraformaldehyde for 30 minutes. The cells were permeabilized using 0.2% Triton X-100, then incubated with cross-reactive goat anti-human SARS-CoV -2-N IgG at 37°C for 1 hour. Afterward, HRP-conjugated goat anti-human IgG (H+L) antibody (1:10000 dilution) (Catalog number: 111-035-144, Jackson ImmunoResearch, West Grove, PA, USA) was added. KPL TrueBlue Peroxidase substrates (Seracare Life Sciences, Inc., Milford, MA, USA) were used to develop the reaction. SARS-CoV-2 foci were quantified using an Elispot reader (Cellular Technology Ltd., Shaker Heights, OH). The viral strains used in this study were isolated from COVID-19 patients in Guangdong, China, including the wild-type (SARS-CoV-2/human/CHN/IQTC01/2020, NCBI accession number: MT123290) and Omicron variants (BA.5, XBB.1, and BQ.1) obtained from the Guangdong Provincial Centre for Disease Control and Prevention, China.

Enzyme-linked immunosorbent assay

Enzyme-linked immunosorbent assay (ELISA) was used to analyze SARS-CoV-2-specific IgG antibodies against the SARS-CoV 2 spike RBD (RBD), spike 1 (S1), and spike 2 (S2) proteins with a commercial antibody detection kit provided by the R&D Department of AtaGenix Laboratories Co., Ltd., Wuhan, China, as previously described.[13] The assay was conducted on plasma samples following the manufacturer's protocol. Absorbance was measured at 450 nm using a BioTek Epoch microplate reader (BioTek Instruments, Inc., Vermont, USA). The IgG concentrations were determined by calculating a standard curve of Ig concentration versus OD450, using the kit's provided standard as reference.

Statistical analysis

Data analysis was carried out using GraphPad Prism version 9.5.1. The Wilcoxon matched-pairs signed rank test was utilized for within-group comparisons across different time points, and the Mann-Whitney test was applied for between-group comparisons. Correlations between variables were examined using Spearman and Pearson methods. The ROC curve was used to assess predictive performance. Moreover, binary logistic regression was used to calculate the combined values for predicting vaccine efficacy. A p value < 0.05 was considered statistically significant.

Result

Inhaled Ad5-nCoV booster induces significantly enhanced and durable neutralizing antibody responses against SARS-CoV-2 variants

To evaluate whether the inhaled Ad5-nCoV booster vaccine effectively induces robust antibody responses, we recruited 35 individuals who had previously received either two (I-I-A, n = 18) or three (I-I-I-A, n = 17) doses of inactivated vaccines,

Table 1. Demographic characteristics.

Group	I-I-A	I-I-I-A	I-I-Bi	I-I-I-Bi
Gender-no.(%)				
Female	12 (66.7%)	13 (76.5%)	23 (69.7%)	37 (46.8%)
male	6 (33.3%)	4 (23.5%)	10 (30.3%)	42 (53.2%)
Age-yrs				
Median	27	26	38	51
Range	22 ~ 49	22 ~ 35	4 ~ 89	20 ~ 84
Interval between second/third dose and booster/infection – days				
Median	212	171	248	302
Range	141-361	54 ~ 263	70-567	139-477

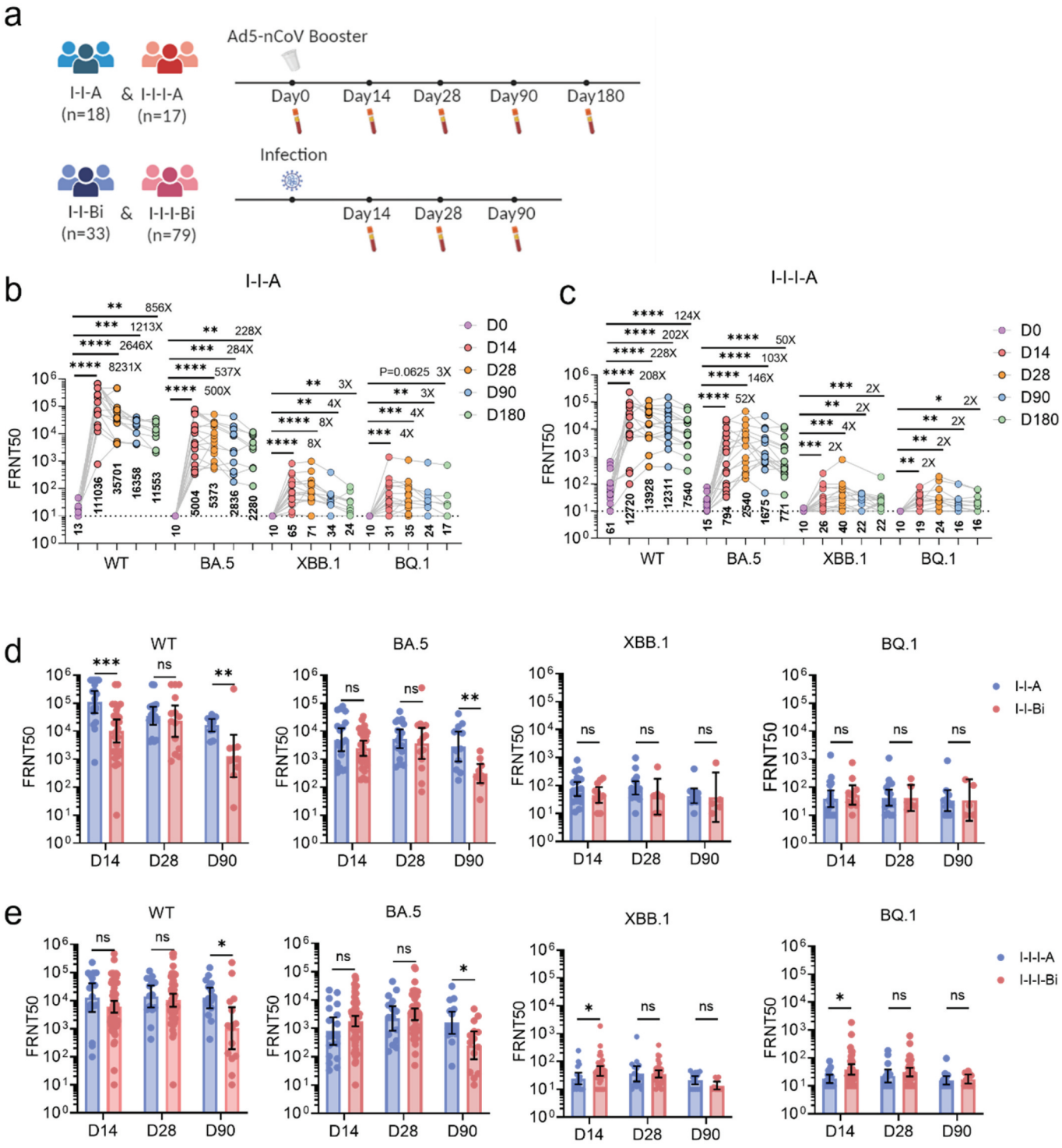

Figure 1. Dynamics of nAb responses against SARS-CoV-2 wild-type (WT) strain and omicron subvariants after aerosolized Ad5-nCoV vaccination following two or three doses of inactivated vaccines. (a) Sampling time points. Created with BioRender.com. (b-c) dynamics of nAb responses of the I-I-A group (b) and the I-I-I-A group (c). The dashed lines represent the detection limit, and the values below the dots stand for the geometric mean of the FRNT50. Wilcoxon matched-pairs signed rank test was performed for comparison. (d–e) comparison of nAb responses induced by sequential aerosolized Ad5-nCoV vaccination and SARS-CoV-2 breakthrough infection. Mann-whitney test was used. ns: $p \geq .5$, *$p < .05$, **$p < .01$, ***$p < .001$, and ****$p < .0001$. titers between different time points.

followed by one booster dose inhaled 2–12 months after the latest intramuscular vaccination. Plasma samples were collected on days 0, 14, 28, 90, and 180 post-booster inhalations (Figure 1a). We then measured the kinetics of neutralizing antibody (nAb) titers against the SARS-CoV-2 wild-type (WT) strain, as well as the BA.5, XBB.1, and BQ.1 variants, using authentic virus neutralization assays.

Group I-I-A showed a greatly significant increase (8231-fold, $p < .0001$) in nAb levels against WT strain at day 14 after vaccination (Figure 1b). Although declined by day 28, it stayed at a durable titer at day 180 after immunization (Figure 1b). Additionally, the nAb titers against BA.5 were exhibited a substantial increase (500-fold, $p < .0001$) at day 14 after immunization as well, which consistently increased until day

28, and maintained half of the nAb levels until day 180. Although the peak antibody responses exhibited a clear escape from XBB.1 and BQ.1 variants, the conserved neutralizing capacities were long-lasting for at least 180 days (Figure 1b). Antibody responses were significantly boosted in group I-I-I-A as well, although with a lower fold-increase compared to group I-I-A (Figure 1c). In addition, group I-I-I-A showed a peak in antibody response to WT, BA.5, XBB.1 and BQ.1 variants at day 28 after immunization, and maintained the neutralizing capacities for over 6 months (Figure 1c).

By better mimicking respiratory infections, aerosolized immunization can more effectively induce respiratory mucosal immune responses compared to intramuscular injection. To determine if the antibody responses from booster mucosal vaccination are comparable to those from breakthrough infections, we included an additional cohort of breakthrough infection cases. This cohort consisted of 33 patients who had completed two doses (I-I-Bi) and 79 who had completed three doses (I-I-I-Bi) of inactivated vaccines before experiencing Omicron breakthrough infections. Plasma samples were collected on days 14, 28, and 90 post-symptom onset (Table 1, Figure 1a).

Regardless of whether participants had a two-dose or three-dose inactivated vaccine background, aerosolized Ad5-nCoV booster vaccination induced neutralizing antibody (nAb) responses comparable to those observed in BA.5 breakthrough infections against both the prototype virus and variants such as BA.5, XBB.1, and BQ.1 (Figure 1d–e). Moreover, at 90 days post the latest immunization, nAb levels against the WT and BA.5 variant were higher in the aerosolized Ad5-nCoV vaccination groups (I-I-A and I-I-I-A) than in the breakthrough infection groups (I-I-Bi and I-I-I-Bi), indicating that aerosolized Ad5-nCoV mounts more durable antibody responses (Figure 1d–e).

Enhanced peak neutralizing antibody response with inhaled aerosol Ad5-nCoV booster following two inactivated vaccine doses compared to three doses

The sequential administration of aerosolized Ad5-nCoV following inactivated vaccination has demonstrated the ability to induce robust antibody responses. However, the optimal timing for this sequential administration remains unclear, necessitating further research to determine the most effective schedule for maximizing the immune response. To address this question, we compared the antibody responses between two booster vaccination groups. Despite the I-I-A group exhibiting lower baseline nAb levels compared to the I-I-I-A group, by day 14 post-booster, the I-I-A group demonstrated significantly higher nAb responses against both the WT strain and omicron variants (Figure 2a, S1). These findings suggest that greater enhancement of antibody responses is achieved when the heterologous aerosolized Ad5-nCoV vaccine is administered as a third dose rather than as a fourth.

To further investigate the differences in antigen targeting profiles of the enhanced antibody responses following heterologous Ad5-nCoV vaccination, we conducted ELISA assays to measure IgG antibodies targeting the S1, S2, and RBD proteins in plasma samples collected on days 0 and 14. The results indicated that IgG antibodies targeting S1 and RBD were predominantly boosted after heterologous Ad5-nCoV inhalation, while those targeting S2 showed only a slight increase (Figure 2b–d). Furthermore, anti-RBD and anti-S1 antibody levels were lower in group I-I-A com-pared to group I-I-I-A on day 0, but were significantly higher on day 14, displaying kinetics similar to the neutralizing antibodies (Figure 2b–c). In contrast, anti-S2 antibody levels were comparable between the two groups before and after booster vaccination (Figure 2d). Consistently, neutralizing antibody levels showed a strong positive correlation with anti-RBD and anti-S1 antibody levels, but not with anti-S2 anti-body levels (Figure 2e–g).

Overall, our results demonstrate that administering the aerosolized Ad5-nCoV vaccine as a booster following two doses of inactivated vaccines results in a stronger enhancement of neutralizing capacities compared to following three doses of inactivated vaccines.

Inverse correlation of prior neutralizing antibody titers with Ad5-nCoV vaccine booster response

Next, we aimed to decipher the contributors to the varied nAb enhancement of Ad5-nCoV vaccination between the I-I-A and I-I-I-A groups. Previous reports have suggested that the enhancement of antibody responses is associated with the intervals between booster vaccinations and preexisting antibody levels before vaccination.[14–17] However, despite a longer interval was observed in group I-I-A than in group I-I-I-A, no significant correlation was found between the boosting intervals and the fold increases of peak nAb levels post-Ad5-nCoV vaccination in either group (Figure S2a–c). When analyzing both groups collectively, a slight but non-significant positive correlation was noted (Figure S2d). This indicates that the boosting interval is not a crucial contributor to the observed differences in antibody enhancement.

Considering that the lower baseline nAb levels are associated with higher peak and greater enhancement after boosting in the I-I-A group (13, 111036, 8231X) com-pared to the I-I-I-A group (61, 13928, 228X) (Figures 1b–c and 2a), we analyzed the association between baseline antibody levels and the enhancement of antibody responses within the two groups. Pearson correlation analysis revealed that the fold-increase in nAb levels following booster Ad5-nCoV inhalation was negatively correlated with the prior nAb levels (Figure 3a), as well as with prior anti-RBD and anti-S1 IgG levels (Figure 3b–c). Conversely, it was positively associated with prior anti-S2 IgG levels (Figure 3d). Similarly, a strong negative correlation was observed between prior antibody titers and the fold-increase in antibody responses for anti-S1, RBD and S2 IgG antibodies (Figure S3a–c). In addition, the ratio of anti-S1 to anti-S2 antibody levels showed a significantly negative correlation with the fold-increase in nAb levels (Figure 3e).

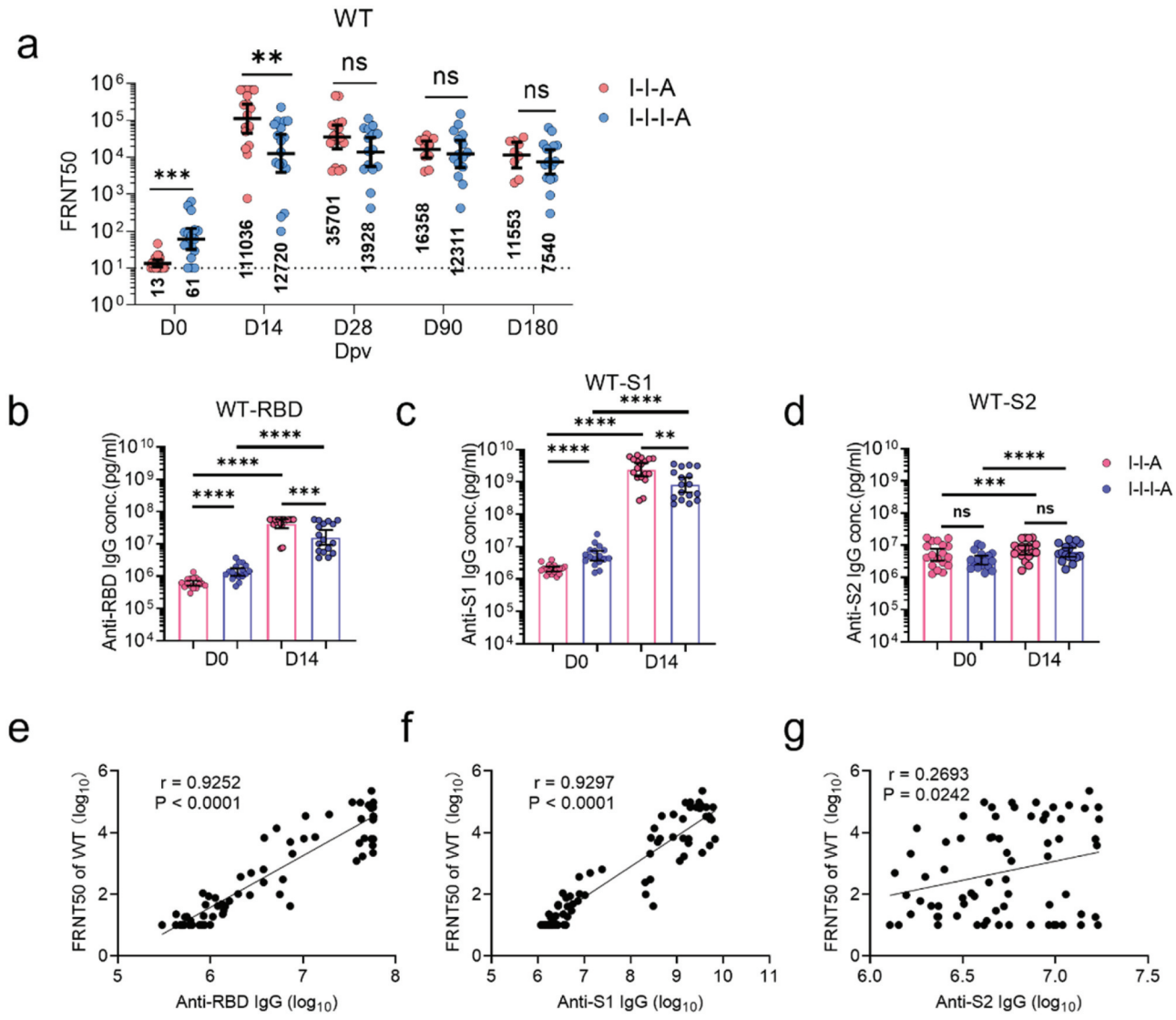

Figure 2. Comparison of antibody responses induced by booster Ad5-CoV inhalation after two- or three-doses of inactivated vaccines. (a) The dynamics of nAb titers against WT strain in the I-I-A group were compared to those in the I-I-I-A group. Dpv: days post-vaccination. (b–d) plasma IgG levels of recognizing the S1, S2 and RBD protein of WT strain in the I-I-A group (red) and the I-I-I-A group (blue). Mann-Whitney test was performed for comparison. ns: $p \geq .5$, *$p < .05$, **$p < .01$, ***$p < .001$ and ****$p < .0001$. (e–g) Pearson correlation analysis was performed between the IgG concentration and the corresponding nAb titers against WT strain.

These results suggest that higher prior nAb titers, accompanied by an antibody response biased toward S1, impair the enhancement of neutralizing capacities following booster aerosolized Ad5-nCoV inhalation.

Pre-existing antibody composition and levels as predictive markers for booster efficacy of aerosolized Ad5-nCoV vaccination

Then we further investigated whether preexisting antibody composition and levels could serve as predictive markers for the efficacy of booster responses to aerosol Ad5-nCoV vaccination. Vaccine recipients were stratified into two subgroups based on strong or weak antibody responses, defined by the fold-increase in nAb titers relative to the geometric mean within groups I-I-A and I-I-I-A. Receiver operating characteristic (ROC) analysis of pre-booster antibody levels were conducted (Figure 4, S4) Result showed that the area under the curve (AUC) of the ROC was 0.6867 for baseline nAb levels, prior anti-RBD IgG of 0.5700, prior anti-S1 IgG of 0.6650 and prior anti-S2 IgG of 0.6667 (Figure 4a, S4a–c). Subsequently, the ratio of anti-S1 to anti-S2 IgG was assessed to integrate the impact of both antibody responses, yielding a higher AUC of 0.7233 (Figure 4b). Combining baseline nAb levels with the anti-S1 to anti-S2 IgG ratio resulted in the highest AUC of 0.7283 (Figure 4c), suggesting a promising predictor of vaccine booster efficacy.

Discussion

Developing effective vaccination strategies and determining optimal booster timing play crucial roles in enhancing and sustaining the immune response for preventing infections from highly prevalent and evolving pathogens like SARS-CoV-2.[5,18] The superiority of heterologous boosters over

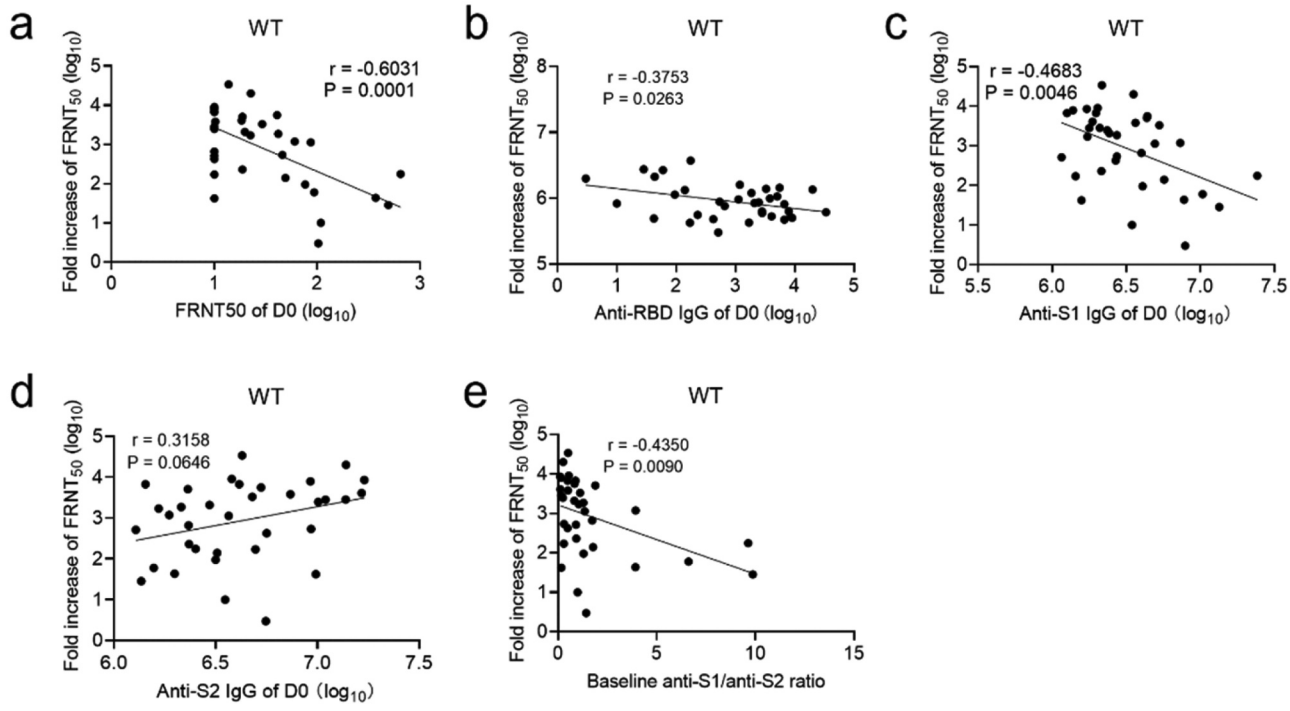

Figure 3. Correlation analysis between the baseline antibody levels and the enhancement of nAb responses induced by Ad5-nCoV vaccination. Pearson correlation analysis was performed between the fold changes of nAb titers and baseline nAb titers (a), the pre-vaccination IgG levels (b–e) against RBD, S1 and S2, and the ratio of anti-S1 to anti-S2 IgG.

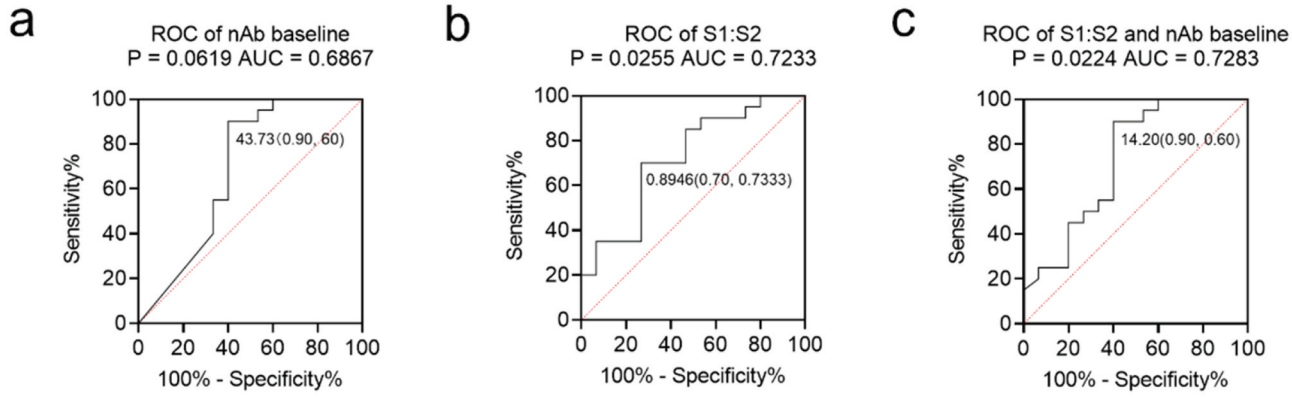

Figure 4. The baseline nAb titers and the ratio of anti-S1 to anti-S2 IgG serve as indicators to predict vaccination efficacy. (a-b) the ROC curve of the nAb baseline (a) and the ratio of anti-S1 to anti-S2 IgG (b) in the vaccination group. AUC means the area under curve and the larger AUC means the better the prediction effect. In the plot, values outside the brackets indicate cutoff values, while those inside indicate paired sensitivity and specificity at these cutoff values. (c) ROC curves were generated within the vaccinated cohort to evaluate the collective predictive capacity of the baseline an-ti-S1 to anti-S2 IgG ratio and the baseline nAb levels. Consider the ratio of anti-S1 IgG to anti-S2 IgG as marker1 and the baseline nAb level as marker2, the cutoff value calculation formula is: marker1+(−0.023)/(−0.075) *marker2.

homologous ones has been established, with aerosolized Ad5-nCoV following two doses of inactivated vaccines showing better outcomes compared to several other injection-based vaccination strategies.[12] However, the impact of varying vaccination backgrounds on antibody response enhancement remains unclear.

In this study, we describe the kinetics of neutralizing antibody response following heterologous boosting with aerosolized Ad5-nCoV after two or three doses of inactivated vaccine. Our data demonstrated that aerosolized Ad5-nCoV inhalation after either two or three doses of inactivated vaccines induced comparable nAb responses against the SARS-CoV-2 WT strain, BA.5, XBB.1, and BQ.1 subvariants, similar to those observed in BA.5 breakthrough infections. The strong and durable nAb response induced by Ad5-nCoV may be attributed to the vaccination route that simulates virus infection and the adenovirus vector.[19,20] These findings suggest that a heterologous booster with aerosolized Ad5-nCoV following two or three doses of inactivated vaccine could be a superior vaccination strategy. However, further data on human efficacy is required for validation.

Interestingly, the I-I-A group exhibited a higher peak nAb response than the I-I-I-A group. Despite a longer interval being observed in the I-I-A group, no significant correlation was found between boosting intervals and the fold increases of peak nAb levels post-Ad5-nCoV vaccination in either group.

This suggests that the boosting interval is likely not a critical contributor to the observed differences in antibody enhancement. Instead, the varied antibody levels in the host prior to the booster vaccination, with antibody titers waning after their peak response to prior vaccination, likely drive this interval influence.

Further investigation revealed that Ad5-nCoV primarily induces peripheral antibodies targeting the S1 epitopes. Prior anti-S1 antibodies were negatively correlated with enhanced nAb, while pre-vaccination anti-S2 antibodies were positively correlated, indicating an epitope-dependent antibody feedback phenomenon. A few studies have reported that higher baseline antibody titers can impair the antibody response following booster vaccination or breakthrough infection.[21,22] Three hypotheses have been proposed to explain this: antibody feedback via inhibition of B cell activation by Fc receptors,[23] antigenic clearance before antigen presentation, and antigen epitope masking by preexisting specific antibodies.[24] However, the antibody feedback mechanism in our study cohort requires further investigation.

Based on these findings, we combined the ratio of pre-vaccination anti-S1 IgG to anti-S2 IgG with baseline nAb levels to predict vaccine efficacy. We found that these factors can serve as reliable predictors of post-vaccination antibody response.

Several limitations exist in this study. First, the present findings are based on a small cohort, thus findings in the study need to be further confirmed in larger cohorts. Second, although respiratory vaccines are superior to intramuscular vaccines in inducing local immune responses and appear to have a stronger capability in blocking infection in animals,[15,25,26] the immune responses in the respiratory tract induced by aerosolized Ad5-nCoV were not assessed in this study because nasal wash or induced sputum sampling was not included in the initial clinical experiment design and was not covered in the ethics application. Moreover, a longer follow-up period can provide a clearer depiction of changes in immune responses following vaccination, offering valuable information for vaccine evaluation and the selection of immunization strategies. However, due to the emergence of new viral strains a year after booster vaccination, most participants experienced breakthrough infections, which hindered the ability to monitor antibody responses over a longer time frame.

Acknowledgments

We sincerely thank all participants in this study. We thank the Biobank for Respiratory Disease at the National Clinical Research Center for Respiratory Disease (BRD-NCRCRD, Guangzhou, Southern China). Ad5-nCoV vaccines were obtained from CanSino Biologics. We thank the Biosafety Level 3 (BSL-3) facilities of Guangzhou Customs District Technology Center.

Disclosure statement

No potential conflict of interest was reported by the author(s).

Funding

Supported by National Natural Science Foundation of China [82025001 JC.Z., 92369113 Y.W., 82172240 Y.W.], R&D Program of Guangzhou Laboratory [EKPG21-30-2 JC.Z.], Guangdong Basic and Applied Basic Research Foundation [2021B1515130005 JX.Z., 2022B1515020059 J.S., 2023B1515020040 Y.W.], and Institute of Immunology in Guangzhou Medical University Funded Project [D18010 JC.Z.].

Notes on contributors

Airu Zhu, currently works in Guangzhou Institute of Respiratory Health, State Key Lab of Respiratory Disease in Guangzhou. Her research interests focus on the host immune responses toward viral infection and vaccination. She has a special interest in infections caused by MERS-CoV, SARS-CoV and SARS-CoV-2 and has extensive experience analyzing T cell and antibody responses in coronavirus-infected mice and humans.

Jincun Zhao, currently works in Guangzhou Institute of Respiratory Health, State Key Lab of Respiratory Disease in Guangzhou. His research interests focus on the relationship between virus replication in the infected host and the immune response to the virus, especially in how the host response to respiratory coronaviruses causes immunopathological disease. He has studied coronaviruses, with an emphasis on pathogenesis for 20 years. He has a special interest in infections caused by MERS-CoV, SARS-CoV and SARS-CoV-2 and has extensive experience analyzing dendritic cell function and T cell and antibody responses in coronavirus-infected mice and humans.

Qihong Yan, currently works in Guangzhou Institute of Respiratory Health, State Key Lab of Respiratory Disease in Guangzhou. His research interests focus on the host immune responses toward viral infection and vaccination. He has a special interest in infections caused by SARS-CoV-2 and Influenza and has extensive experience analyzing immune repertoire data.

Author contributions

Conceptualization, A.Z. and JC.Z.; Methodology, C.C., Z.C., Y.W., A. Z. and JC.Z.; Investigation, C.C., T.T., Z.C., L.C., J.C., F.L., J.S., JX.Z., Y. W., Q.Y., A.Z.; Validation: C.C., T.T., Z.C., L.C., J.C.; Formal analysis, C. C., Z.C. and A.Z.; Resources, T.T., L.C., J.C., Z.X., F.L., J.S., Q.Y.; Data curation, C.C., A.Z.; Writing original draft preparation, C.C., Z.C.; Writing review and editing, A.Z. and JC.Z.; Visualization, C.C., Z. C. and A.Z.; Supervision, Q.Y., A.Z., and JC.Z.; Project administration, A.Z. and JC.Z.; Funding acquisition, Y.W., J.S., JX.Z. and JC.Z.; All authors have read and agreed to the published version of the manuscript.

Data availability statement

The data of this study can be obtained from this article and its supplementary materials.

Ethics approval and consent to participate

The study was approved by the Medical Ethics Committee of the First Affiliated Hospital of Guangzhou Medical University [No 2022-G-42]. All participants have given informed consent to participate voluntarily and confidentially in the study.

References

1. Tregoning JS, Flight KE, Higham SL, Wang Z, Pierce BF. Progress of the COVID-19 vaccine effort: viruses, vaccines and variants versus efficacy, effectiveness and escape. Nat Rev Immunol. 2021;21(10):626–636. doi:10.1038/s41577-021-00592-1.
2. Mohamed Khosroshahi L, Rokni M, Mokhtari T, Noorbakhsh F. Immunology, immunopathogenesis and immunotherapeutics of COVID-19; an overview. Int Immunopharmacol. 2021;93:107364. doi:10.1016/j.intimp.2020.107364.

3. COVID-19 vaccine tracker and landscape. [accessed 2023 Mar 30] Available from: https://www.who.int/publications/m/item/draft-landscape-of-covid-19-candidate-vaccines.
4. Hassan AO, Kafai NM, Dmitriev IP, Fox JM, Smith BK, Harvey IB, Chen RE, Winkler ES, Wessel AW, Case JB, et al. A single-dose intranasal ChAd vaccine protects upper and lower respiratory tracts against SARS-CoV-2. Cell. 2020;183(1):169–184.e113. doi:10.1016/j.cell.2020.08.026.
5. Waltz E. How nasal-spray vaccines could change the pandemic. Nature. 2022;609(7926):240–242. doi:10.1038/d41586-022-02824-3.
6. Afkhami S, D'Agostino MR, Zhang A, Stacey HD, Marzok A, Kang A, Singh R, Bavananthasivam J, Ye G, Luo X, et al. Respiratory mucosal delivery of next-generation COVID-19 vaccine provides robust protection against both ancestral and variant strains of SARS-CoV-2. Cell. 2022;185(5):896–915.e819. doi:10.1016/j.cell.2022.02.005.
7. Langel SN, Johnson S, Martinez CI, Tedjakusuma SN, Peinovich N, Dora EG, Kuehl PJ, Irshad H, Barrett EG, Werts AD, et al. Adenovirus type 5 SARS-CoV-2 vaccines delivered orally or intranasally reduced disease severity and transmission in a hamster model. Sci Transl Med. 2022;14(658):eabn6868. doi:10.1126/scitranslmed.abn6868.
8. Wu S, Huang J, Zhang Z, Wu J, Zhang J, Hu H, Zhu T, Zhang J, Luo L, Fan P, et al. Safety, tolerability, and immunogenicity of an aerosolised adenovirus type-5 vector-based COVID-19 vaccine (Ad5-nCoV) in adults: preliminary report of an open-label and randomised phase 1 clinical trial. Lancet Infect Dis. 2021;21(12):1654–1664. doi:10.1016/S1473-3099(21)00396-0.
9. Chen J, Wang P, Yuan L, Zhang L, Zhang L, Zhao H, Chen C, Wang X, Han J, Chen Y, et al. A live attenuated virus-based intranasal COVID-19 vaccine provides rapid, prolonged, and broad protection against SARS-CoV-2. Sci Bull (Beijing). 2022;67(13):1372–1387. doi:10.1016/j.scib.2022.05.018.
10. Li JX, Wu S-P, Guo X-L, Tang R, Huang B-Y, Chen X-Q, Chen Y, Hou L-H, Liu J-X, Zhong J, et al. Safety and immunogenicity of heterologous boost immunisation with an orally administered aerosolised Ad5-nCoV after two-dose priming with an inactivated SARS-CoV-2 vaccine in Chinese adults: a randomised, open-label, single-centre trial. Lancet Respir Med. 2022;10(8):739–748. doi:10.1016/S2213-2600(22)00087-X.
11. Zhong J, Liu S, Cui T, Li J, Zhu F, Zhong N, Huang W, Zhao Z, Wang Z. Heterologous booster with inhaled adenovirus vector COVID-19 vaccine generated more neutralizing antibodies against different SARS-CoV-2 variants. Emerg Microbes Infect. 2022;11(1):2689–2697. doi:10.1080/22221751.2022.2132881.
12. Zhu A, Wei P, Man M, Liu X, Ji T, Chen J, Chen C, Huo J, Wang Y, Zhao J, et al. Antigenic characterization of SARS-CoV-2 omicron subvariants XBB.1.5, BQ.1, BQ.1.1, BF.7 and BA.2.75.2. Signal Transduct Target Ther. 2023;8(1):125. doi:10.1038/s41392-023-01391-x.
13. Qiu S, Chen Z, Zhu A, Zeng Q, Liu H, Liu X, Ye F, Jin Y, Wu J, Yang C, et al. Successful clearance of persistent SARS-CoV-2 asymptomatic infection following a single dose of Ad5-nCoV vaccine. Signal Transduct Targeted Ther. 2023;8(1). doi:10.1038/s41392-023-01345-3.
14. Dangi T, Sanchez S, Lew MH, Awakoaiye B, Visvabharathy L, Richner JM, Koralnik IJ, Penaloza-MacMaster P. Pre-existing immunity modulates responses to mRNA boosters. Cell Rep. 2023;42(3):112167. doi:10.1016/j.celrep.2023.112167.
15. Murray SM, Ansari AM, Frater J, Klenerman P, Dunachie S, Barnes E, Ogbe A. The impact of pre-existing cross-reactive immunity on SARS-CoV-2 infection and vaccine responses. Nat Rev Immunol. 2023;23(5):304–316. doi:10.1038/s41577-022-00809-x.
16. Schaefer-Babajew D, Wang Z, Muecksch F, Cho A, Loewe M, Cipolla M, Raspe R, Johnson B, Canis M, DaSilva J, et al. Antibody feedback regulates immune memory after SARS-CoV-2 mRNA vaccination. Nature. 2022;613(7945):735–742. doi:10.1038/s41586-022-05609-w.
17. Miyamoto S, Arashiro T, Adachi Y, Moriyama S, Kinoshita H, Kanno T, Saito S, Katano H, Iida S, Ainai A, et al. Vaccination-infection interval determines cross-neutralization potency to SARS-CoV-2 omicron after breakthrough infection by other variants. Med. 2022;3(4):249–261.e244. doi:10.1016/j.medj.2022.02.006.
18. Chi WY, Li Y-D, Huang H-C, Chan TEH, Chow S-Y, Su J-H, Ferrall L, Hung C-F, Wu T-C. COVID-19 vaccine update: vaccine effectiveness, SARS-CoV-2 variants, boosters, adverse effects, and immune correlates of protection. J Biomed Sci. 2022;29(1):82. doi:10.1186/s12929-022-00853-8.
19. Zhang L, Jiang Y, He J, Chen J, Qi R, Yuan L, Shao T, Zhao H, Chen C, Chen Y, et al. Intranasal influenza-vectored COVID-19 vaccine restrains the SARS-CoV-2 inflammatory response in hamsters. Nat Commun. 2023;14(1). doi:10.1038/s41467-023-39560-9.
20. Zhang Y, Liu J, Li H, Yuan F, Jiang C, Cang T, Li K, Hu Q, Liu J, Zheng A, et al. Comparison of the immunogenicity of nasal-spray rVSV vector, adenovirus vector, and inactivated Covid-19-based vaccines in rodent models. J Med Virol. 2023;95(5). doi:10.1002/jmv.28806.
21. Das S, Singh J, Shaman H, Singh B, Anantharaj A, Sharanabasava P, Pandey R, Lodha R, Pandey AK, Medigeshi GR, et al. Pre-existing antibody levels negatively correlate with antibody titers after a single dose of BBV152 vaccination. Nat Commun. 2022;13(1). doi:10.1038/s41467-022-31170-1.
22. Gao B, He L, Bao Y, Chen Y, Lu G, Zhang Y, Xu Y, Su B, Xu J, Wang Y, et al. Repeated vaccination of inactivated SARS-CoV-2 vaccine dampens neutralizing antibodies against omicron variants in breakthrough infection. Cell Res. 2023;33(3):258–261. doi:10.1038/s41422-023-00781-8.
23. Amigorena S, Bonnerot C, Drake JR, Choquet D, Hunziker W, Guillet J-G, Webster P, Sautes C, Mellman I, Fridman WH, et al. Cytoplasmic domain heterogeneity and functions of IgG Fc receptors in B lymphocytes. Science. 1992;256(5065):1808–1812. doi:10.1126/science.1535455.
24. Heyman B. Antibody feedback suppression: towards a unifying concept? Immunol Lett. 1999;68(1):41–45. doi:10.1016/S0165-2478(99)00028-0.
25. Lei H, Alu A, Yang J, He X, He C, Ren W, Chen Z, Hong W, Chen L, He X, et al. Cationic crosslinked carbon dots-adjuvanted intranasal vaccine induces protective immunity against omicron-included SARS-CoV-2 variants. Nat Commun. 2023;14(1). doi:10.1038/s41467-023-38066-8.
26. Fraser R, Orta-Resendiz A, Mazein A, Dockrell DH. Upper respiratory tract mucosal immunity for SARS-CoV-2 vaccines. Trends Mol Med. 2023;29(4):255–267. doi:10.1016/j.molmed.2023.01.003.