


















ORIGINAL RESEARCH



Antibody response to the first dose of AZD1222 vaccine in COVID-19 convalescent and uninfected individuals in Bangladesh

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ABSTRACT

Background: Vaccination with the Oxford-AstraZeneca COVID-19 vaccine (AZD1222) initially started in the UK and quickly implemented around the Globe, including Bangladesh. Up to date, more than nine million doses administered to the Bangladeshi public.

Method: Herein, we studied the antibody response to the first dose of AZD1222 in 86 Bangladeshi individuals using in-house ELISA kits. Study subjects were categorized into two groups, convalescent and uninfected, based on prior infection history and SARS-CoV-2 nucleocapsid-IgG profiles.

Results: All the convalescent individuals presented elevated spike-1-IgG compared to 90% of uninfected ones after the first dose. Day >28 post-vaccination, the convalescent group showed six times higher antibody titer than the uninfected ones. The most elevated antibody titers for the former and later group were found at Day 14 and Days >28 post-vaccination, respectively. The spike-1-IgA titer showed a similar pattern as spike-1-IgG, although in a low-titer. In contrast, the IgM titer did not show any significant change in either group.

Conclusion: High antibody titer in the convalescent group, signify the importance of the first dose among the uninfected group. This study advocates the integration of antibody tests in vaccination programs in the healthcare system for maximizing benefit.

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1. Introduction

Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of the coronavirus disease-2019 (COVID-19) pandemic, has affected over 185 million people with 4 million deaths globally [1]. Unavailability of an experimental or proven anti-viral drug leads the physician to manage COVID-19 patients through symptomatic management [2–4]. Over the last few decades, advances in bioprocess technology and vaccine research have introduced technologies such as mRNA or vector-based vaccine technologies over the traditional whole live/inactivated vaccines [5]. Both mRNA and vector-based COVID-19 vaccines have shown promising results, albeit varying degrees of efficiencies and significantly lower death rates after vaccination [6]. Although rare side effects such as blood-clot, cerebral venous sinus thrombosis

with thrombocytopenia (TTS), or anaphylaxis has been reported with the administration of Oxford-AstraZeneca (AZD1222), Johnson & Johnson (J&J), Pfizer, and Moderna vaccines stopping the rampage of this pandemic largely depend upon successful vaccination of majority of the population within a short period [7–9].

Complete immune protection against SARS-CoV-2 requires the efficient and coordinated action of the dual-faceted swords of the human immune response, i.e. humoral and cell-mediated immunity [10]. During natural infection with SARS-CoV-2 human body produces neutralizing antibodies (IgG) against various regions within the virus's spike (S) protein. Voss *et al.* has recently reported that >80% of antibodies developed after natural infection targets spike protein regions outside the receptor-binding domain (RBD) but have

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 Supplemental data for this article can be accessed [here](#)

Article Highlights

- The total (100%) of the convalescent subjects were seroconverted after the first dose of AZD1222.
- About 90% of the subjects with no COVID-19 history prior vaccination seroconverted after the first dose of AZD1222.
- The first dose of the AZD1222 vaccine work as a booster dose for the subjects with previous infection history before immunization.
- About 10% of subjects without prior infection history become Rt-qPCR positive after 28 days of the first dose of AZD1222 vaccination but recovered quickly.
- IgG profile of SARS-CoV-2 specific nucleocapsid can differentiate between infection status.
- Along with S1-IgG, S1-IgA can also be used as a serological marker of seroconversion post-vaccination.

neutralizing capacity [11]. In contrast, the significance of antibodies against epitopes other than spike is yet to be unrevealed but may confer some cross-protection [12]. Even though neutralizing antibodies (S-IgGs) does not always make an individual resistant to the virus or lessen COVID-19 disease severity, the antibody can exhibit a sustained response over 6–8 months following infection [13–15]. While humoral immunity is mainly measured through IgG, the early antibody-mediated neutralizing response is dominated by IgA [16]. Knowledge on neutralizing antibodies in COVID-19 convalescent individuals is essential to understand the protective immunity against reinfection and the state of herd immunity of a nation.

The vector-based or mRNA COVID-19 vaccines have been designed to provoke an immune response against the SARS-CoV-2 S protein. The AZD1222 vaccine-induced seroconversion in 97.1% of the healthcare workers, among 890 subjects studied while observing a higher IgG level in the convalescent group [6]. The observation of high seroconversion among the vaccinated groups ultimately led to Emergency Use Authorization (EUA) by January 2021 in the UK and other countries [17]. A recent UK population-based study on the impact of the first dose of the AZD1222 vaccine found an almost 65% reduction in SARS-CoV-2 cases [18]. Nevertheless, the vaccine has been reported to elicit T and B cell responses after a single dose [19,20].

In February 2021, Bangladesh launched the COVID-19 vaccination program with the AZD1222 produced in India's Serum Institute of India (SII). The detection rate for COVID-19 in Bangladesh remained significantly low considering its population density; however, a plausible reason for that could be insufficient testing [21,22]. Moreover, unlike other countries, no serological kit has been approved for COVID-19 diagnostic purposes, leaving the knowledge of the epidemiological extent of infection incomplete. Ethnic variability, epigenetic and environmental factors, and socio-demographic behavior can affect the efficiencies of a vaccination program [23,24]. Since there were no third phase pre-clinical trials for any vaccine in Bangladesh, data on antibody response after vaccination against SARS-CoV-2 is also missing.

This study was designed to understand the humoral-immunity response to the first dose of the AZD1222 vaccine in COVID-19 convalescent and uninfected individuals in

Bangladesh. Our observations coincide with UK studies of the AZD1222 vaccine and other vaccines in place. It was found to be more effective in spiking antibody titer among pre-infected ones than the naïve population. The observation of this work is expected to imply and improve the current COVID-19 vaccination program and the future pandemic vaccine development research database.

2. Materials and methods

2.1. Subject enrollment

Our cross-sectional study included patients of varied backgrounds. We collected data from front-line workers and others. Participants who planned to take the vaccine were invited through the social media platform. Serum samples were collected from the participants who agreed and provided their consent for participation in the study. Samples were collected at different times points, based on the subjects' availability. The collection period was stratified into before vaccination and post-vaccination periods. Furthermore, the post-vaccination period was divided into four sub-classes, i.e. day 0–7, day 7–14, day 14–28, and day>28 (day 29–56).

2.2. Classification of subjects

Subjects were classified into two major groups based on COVID-19 infection before vaccination. Those who had COVID-19 like symptoms, confirmed by RT-qPCR, and positive for IgG against nucleocapsid (NCP), denoted as the 'Convalescent' ones. Those who did not have any clinical symptoms and signs or were negative in RT-qPCR testing and nucleocapsid (NCP) IgG were denoted as the 'Uninfected' ones.

2.3. Antibody profiling and characterization of antibody dynamics

Previously, we developed in-house ELISA assays to characterize IgG, IgM, and IgA antibodies against SARS-CoV-2 specific NCP, S1, RBD, and S1+ S2 antigens [25]. In this study, four in-house assays were used, namely, ELISA assay for detecting IgG, IgM, and IgA specific for S1 and assay for detecting IgG against NCP [26,27]. Patient samples were grouped into convalescent and uninfected groups by employing the NCP-IgG assay and previous infection history. The ELISA assay plates were coated with 1:100–200 dilution of NCP and S1 antigens (SinoBiologicals, China). After blocking, the plates were inoculated with diluted (1:100) samples for 15 minutes. Next, 1:4000, 1:3000, and 1:2500 dilutions of horseradish peroxidase (HRP) conjugated anti-human IgG (The NativeAntigens, UK), anti-human IgM (The NativeAntigens, UK), and anti-human IgA (Abcam, UK) were added to each well for the respective assays. Finally, 3,3',5,5'-Tetramethylbenzidine (TMB) was added as substrate, followed by a stop solution. The reaction was read at 450 nm wavelength. Previously confirmed and well-characterized samples were used as positive and negative controls [26,27].

2.4. Ethical approval

The experiment was conducted as per the 1964 Helsinki constitutions. Institutional ethical approval was provided by the Institutional Review Boards of Jahangirnagar University (BBEC, JU/M2021/COVID-19/5(2)). Written consent forms and questionnaires were provided to the subjects. The subjects have explained the purpose of the study, and samples were withdrawn after proper consenting.

2.5. Statistical analysis

Data were presented as either mean, standard deviation, median with inter-quartile range, frequencies, or percentages. Bivariate association between convalescent and uninfected and outcomes (NCP-IgG, S1-IgA, S1-IgM, and S1-IgG) were assessed using either Spearman's rank correlation, Mann-Whitney U-test, or Kruskal-Wallis. The mean difference between convalescent and uninfected with NCP-IgG, S1-IgA, S1-IgM, and S1-IgG were estimated using an independent sample *t*-test or univariate regression model. A *p*-value of <0.05 was considered significant. The statistical analyses were performed using Stata 13 (StataCorp, LP, College Station, Texas, USA) and SPSS (version 22.0), and the graphical presentation was made using GraphPad Prism (version 8.3.1).

3. Results

A total of 86 participants were enrolled in this study, from whom a total of 138 samples were collected and analyzed. The mean age of the registered participants was 44 years (SD \pm 13.6). Among the participants, two males and one female, who were previously uninfected, became infected with SARS-CoV-2 after Day 28 from their first dose of the AZD1222 vaccine and were excluded from the study. Finally, fifty-four (65.1%) male participants, while twenty-nine (34.9%) female participants were enrolled in the study (*n* = 83) (Table 1).

It is well documented that previously infected COVID-19 patients develop IgG antibodies against NCP and S1 proteins

[28]. Eighty-three participants were further divided into two groups based on their NCP-IgG antibody levels and history of RT-qPCR test to eliminate the active infection. Among the 83 participants, 50 had a previous history of SARS-CoV-2 infection (RT-qPCR positive minimum 56 days before vaccination), while 33 participants were either without any history of exposure or RT-qPCR and anti-NCP-IgG negative (Table 1; Supplementary Table 1). We observed that the mean average of NCP-IgG in the uninfected group was below the cutoff range for all five-time points (Before vaccination: 0.82 ± 0.13 ; Day 7: 0.76 ± 0.20 ; Day 14: 0.90 ± 0.24 ; Day 28: 0.83 ± 0.26 ; and Day >28: 1.00 ± 0.66) (Figure 1; Supplementary Table 1). On the other hand, mean averages of NCP-IgG in the convalescent group remained higher than the cutoff range (Before vaccination: 2.02 ± 1.62 ; Day 7: 2.10 ± 1.28 ; Day 14: 1.88 ± 1.58 ; Day 28: 1.38 ± 0.83 ; and Day >28: 2.01 ± 0.87) (Supplementary Table 1). Before vaccination, the NCP-IgG value was significantly higher, 1.20 times (95% CI, 0.28, 2.11; *p* = 0.012), in convalescent participants than uninfected groups. Similar pattern was observed at Day 7 (*p* = 0.007) and Day >28 (*p* = 0.009) (Figure 1; Supplementary Table 1).

In the case of the S1-IgG antibody, the convalescent group had 3.45 times higher (95% CI: 1.47, 5.46; *p* = 0.001) antibody titer than the uninfected control group before vaccination (Figure 2; Supplementary Table 2). The mean S1-IgG average in the convalescent group were 4.58 ± 3.48 before vaccination, 10.67 ± 4.10 , 12.19 ± 4.12 , 11.38 ± 3.05 , and 10.59 ± 3.64 compared to 1.11 ± 0.79 , 1.66 ± 0.91 , 2.90 ± 2.36 , 1.15 ± 0.63 , and 4.48 ± 3.70 in an uninfected group before vaccination, at 7, 14, 28, and >28 days, respectively (Figure 2; Supplementary Table 2). The previously infected participants showed a continuous increase of OD/cutoff titers from Day 7 to Day28 while the differences remained significant (Figure 2). Interestingly, at >28 days, the level of S1-IgG decreased but remained 6.10 times (95% CI 2.95, 9.26; *p* = 0.001) higher than uninfected participants (Figure 2; Supplementary Table 2).

Before vaccination, there was no significant difference in S1-IgA and S1-IgM levels between convalescent and uninfected individuals (Figure 3 and Supplementary Figure 1).

Table 1. Socio-demographic and clinical characteristics of the study participants.

Variables	Overall (n = 83)	Convalescent (n = 50)	Uninfected (n = 33)
Age, years	44.46 \pm 13.63	47.02 \pm 14.34	40.58 \pm 11.66
Gender			
Male	54(65.1%)	33(66.0%)	21(63.6%)
Female	29(34.9%)	17(34.0%)	12(36.4%)
Occupation			
Service	41(49.4%)	21(42.0%)	20(60.6%)
Housewife	9(10.8%)	6(12.0%)	3(9.1%)
Health worker	12(14.5%)	9(18.0%)	3(9.1%)
Unemployed	16(19.3%)	10(20.0%)	6(18.2%)
Business	5(6.0%)	4(8.0%)	1(3.0%)
Monthly income, taka			
<15,000	28(34.0%)	17(34.0%)	11(33.3%)
15,000–65,000	33(39.8%)	18(36.0%)	15(45.5%)
>65,000	22(26.5%)	15(30.0%)	7(21.2%)
Education in years			
<14	28(33.7%)	19(38.0%)	9(33.7%)
15–18	38(45.8%)	22(44.0%)	16(48.5%)
>18	17(20.5%)	9(18.0%)	8(24.2%)
*H/O comorbidities	15(18.1%)	12(24.0%)	3(9.1%)

Data were presented as mean \pm Std or number with percent in the parenthesis. H/O = History Of.

*The patients had either hypertension or CKD or Diabetes or heart problem or cancer

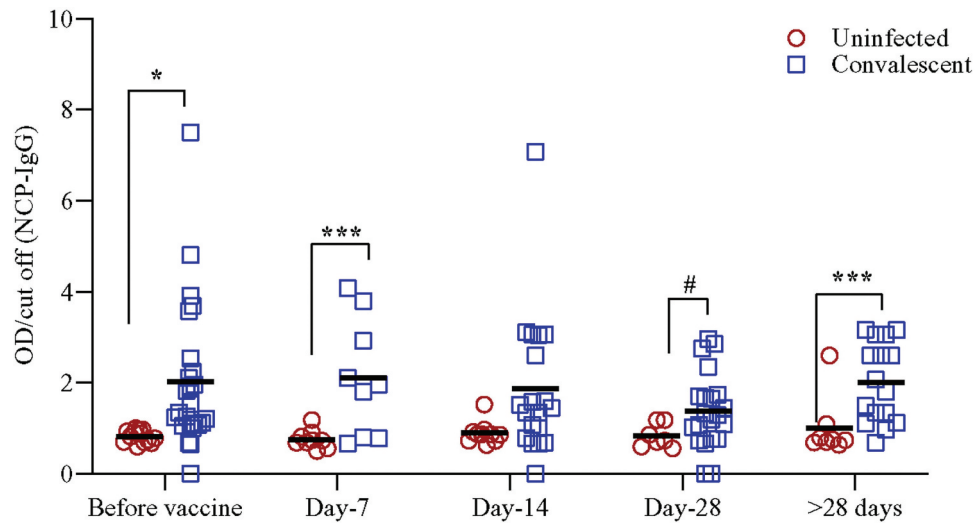


Figure 1. SARS-CoV-2 anti-nucleocapsid IgG (NCP-IgG) level in convalescent and uninfected vaccinated participants. Throughout the study, the participants did not present an increase in antibodies against NCP protein. However, there was a significant difference in the antibody titer among the uninfected and convalescent participants. Moreover, after vaccination, the overall trend remains that the convalescent participants of the NCP-IgG decreased, though they remained steady throughout the study. An asterisk represents a significant difference in NCP-IgG between uninfected and convalescent subjects (* <0.05, **<0.01, and ***<0.001) using an Independent Sample t-test.

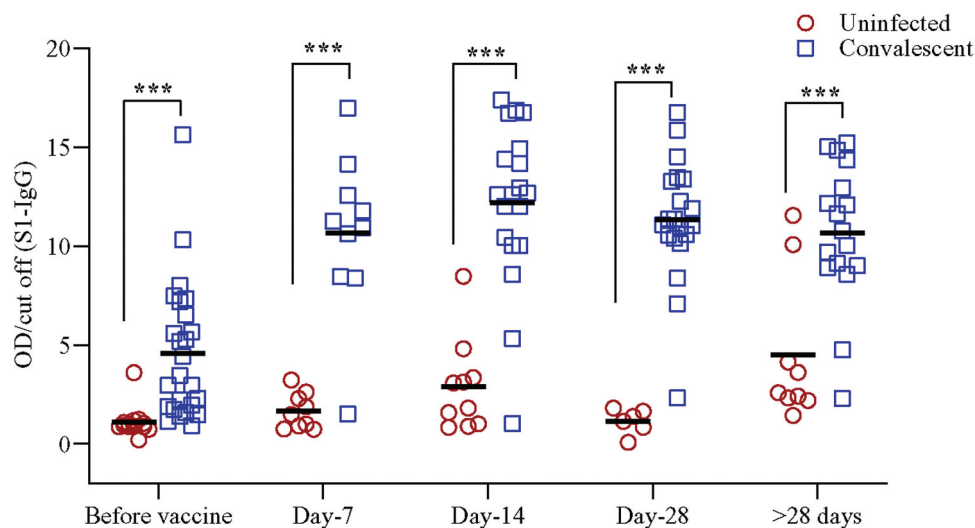


Figure 2. SARS-CoV-2 anti-spike-1 IgG (S1-IgG) development in convalescent and uninfected participants before and after vaccination. There was a significant difference in the development of antibody levels among the uninfected and convalescent participants. However, the development of S1-IgG in convalescent subjects reached its highest titer within 14 days post-vaccination, whereas, for the uninfected group, it reached the level similar to that of the convalescent group before vaccination. An asterisk represents a significant difference in S1-IgG between uninfected and convalescent subjects (* <0.05, **<0.01, and ***<0.001) using an Independent Sample t-test.

However, S1-IgM was significantly higher after seven days post-vaccination in convalescent participants (Supplementary Figure 1). Whereas, at day-7, -14, -28, and >28, the S1-IgA value remained 1.34, 2.54, 2.78, and 1.75 times higher, respectively, in convalescent participants, and the differences were significant (Figure 3 and Supplementary Table 3).

4. Discussion

This research found a significant difference in the S1-IgG level between individuals with previous SARS-CoV-2 infection and uninfected individuals before vaccination (Figures 2, 4 and 5). We observed 100% seroconversion in the convalescent group,

whereas 90% in the uninfected group, after the first dose of AZD1222 vaccine. Furthermore, more than 28 days post-vaccination, the former group developed about six times higher antibody titer compared to the uninfected ones (Figures 2, 4; Supplementary Table 2). A similar observation was found in a study conducted on healthcare workers in the UK, where after administering the first dose of Pfizer-BioNtech and AZD1222 vaccine, 2706/2720 (99.5%) and 864/890 (97.1%), respectively, were seroconverted. Moreover, higher antibody titers were found in the convalescent group than the previously uninfected one [6]. The antibody generated after the first dose of vaccination in the convalescent group is comparable to that of uninfected individuals receiving two

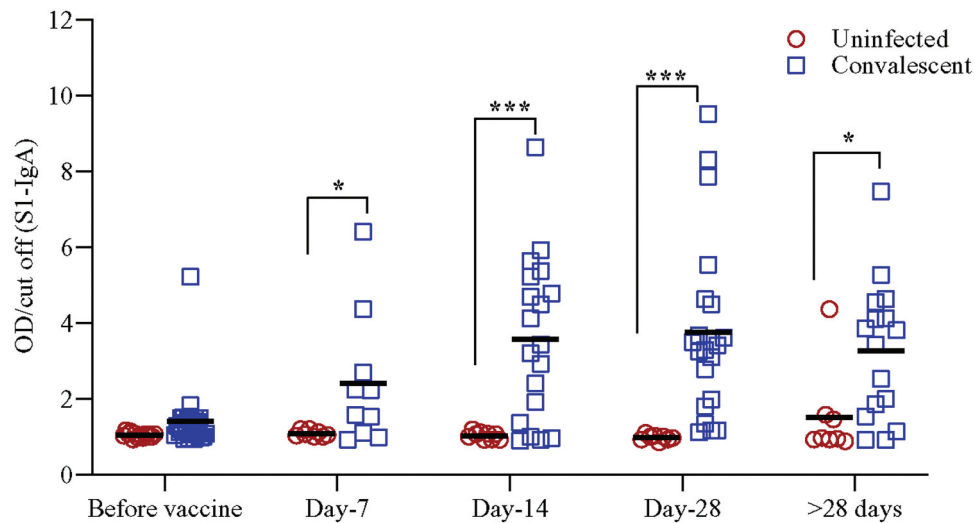


Figure 3. SARS-CoV-2 anti-spike-1 IgA (S1-IgA) development in participants before and after vaccination. There was a significant increase in the antibody titer among both the uninfected and convalescent groups. However, after Day >28, an uninfected group of participants presented a higher antibody titer level than the previous collection. An asterisk represents a significant difference in S1-IgA between uninfected and convalescent subjects (* <0.05, **<0.01, and ***<0.001) using an Independent Sample t-test.

doses of vaccination (Figure 5) [29–31]. Sadat et al. found that, after a single dose of immunization of 56 volunteers with Pfizer-BioNTech or Moderna vaccine, a classical secondary response was observed in the convalescent group [31]. On the other hand, Wei et al. reported lower antibody development in older people, male and comorbid ones after the first dose of AZD1222 and Pfizer-BioNTech vaccine [32]. However, no such association was noted in this study.

The COVID-19 infected patients generally develop antibodies against both NCP and S1 antigens, where the S1 antibody is known to neutralize the virus [33,34]. In the real-world situation, determining the immediate efficacy of the vaccine, measuring the neutralizing antibody spiking is one of the key ways. Therefore, the study subjects must be differentiated among the group with or without prior infection history to interpret the result better [35]. Though memory B-cell or T-cell can give the actual insight of this status (Figure 5), NCP-IgG can be the serological marker of the previous infection, as long the antibodies do not wane off [34]. Among the convalescent group, twelve post-COVID-19 subjects gave blood multiple times (2–4 times). This study observed a gradual decline of NCP-IgG over time among this sub-group (Supplementary Table 1). COVID-19 infection after vaccination has been an often-reported phenomenon [35,36]. We found that three participants from the uninfected group become RT-qPCR positive after around 28 days post-vaccination. The participants were excluded from our panel to avoid any confusion. Interestingly, >28 days post-vaccination of the uninfected group showed the NCP-IgG mean average of 1.00 ± 0.66 , indicating that of the eight participants, some of them may have developed asymptomatic infection post-vaccination and presented elevated NCP-IgG (Figure 1; Supplementary Table 1). We identified one uninfected participant who didn't report any signs and symptoms but showed a gradual increase in NCP-IgG (Figure 1). This subject may have been an asymptomatic case who was infected with SARS-CoV-2 post-vaccination.

The Spike protein sequence used in AZD1222 is derived from the early strains. Diagnostic assay kits developed using a similar peptide sequence, expressed in a mammalian cell line, would represent the epitopes in a conformation identical to that of natural SARS-CoV-2 [37,38]. Moreover, recombinant spike proteins generally have the neutralizing capacity, i.e. they would compete with natural/pseudovirus in ACE-2 binding [39]. We tried to cover these issues by utilizing an ACE-2 binding protein that can successfully neutralize the SARS-CoV-2 pseudovirus [40]. Thus, post-vaccination, an increase of antibodies specific to S antigens can be evaluated with this antigen. Moreover, antibodies against the S2 region of SARS-CoV-2 can cross-react with other coronaviruses. Thus the S1 subunit was chosen for this study [41].

This study observed the enhancement of vaccine-specific IgG antibodies, and the current observation was in the same line as the earlier researches [6,29,30]. We found that subjects with prior infection history have significantly higher IgG antibody titer than uninfected ones (Figures 2, 4, 5; Supplementary Table 2). In the study group, three uninfected subjects did not seroconvert, even after 28 days, leaving the seroconversion percentage for this group at ~90%. It is interesting to see whether the seronegative subjects become seropositive after the second dose in our future endeavors. In contrast, two participants from this group showed seroconversion levels similar to that of the convalescent group at days >28 (Figure 2). One of these participants reported higher NCP-IgG, and both were probably exposed to SARS-CoV-2 post-vaccination without developing any symptoms. Interestingly, in the convalescent group, two subjects didn't seroconvert either (Figure 2). This can be explained by the reports showing that a fraction of the population may not develop antibodies after natural Covid-19 infection or vaccination but have T-cell mediated immunity [42–44].

The first dose of vaccine work as a booster dose for the convalescent group and the highest titer was observed between 7–14 days post-vaccination. This occurrence was

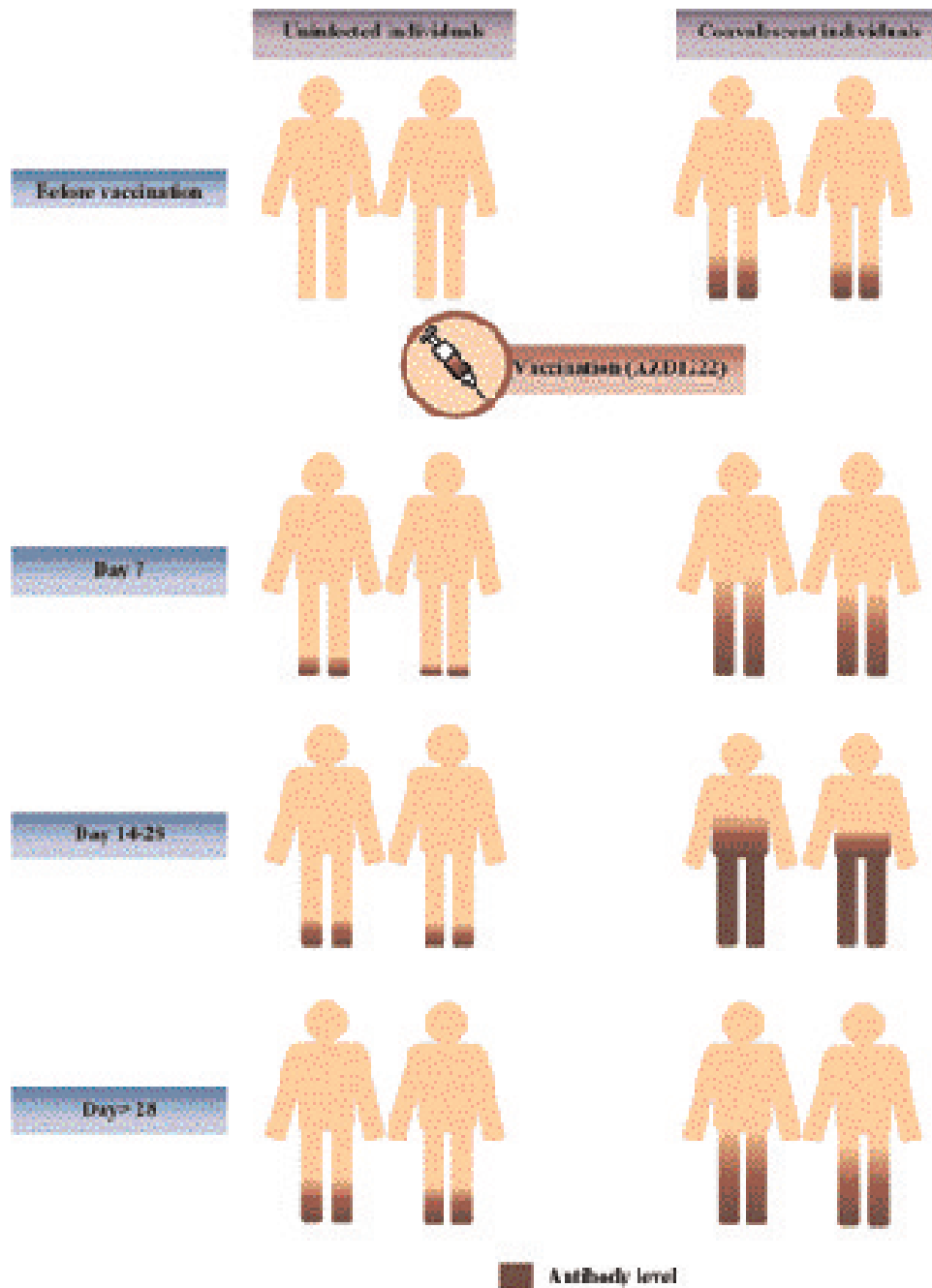


Figure 4. Antibody levels of two groups of individuals with (Right) and without (Left) prior SARS-CoV-2 infection. The antibody response was higher and faster in individuals who have been previously diagnosed with COVID-19 infection. However, after 28 days, the antibody level flattens out but stays at a higher titer. In contrast, individuals who have not been previously exposed to SARS-CoV-2 present a slower increase of antibody titers, which continues beyond 28 days.

previously observed in researches on other vaccine candidates as well [40]. However, the titer increased slowly for the uninfected group and reached the highest level >28 days after vaccination, but remained about one-sixth of the titer of the convalescent group (Figures 2 and 4).

The circulating plasmablasts in the blood express IgA1 specific against SARS-CoV-2 spike protein a few days before IgG comes into play [16,25,45,46]. These IgA1 have virus neutralizing capacity and should be used as serological markers along with IgG. Here, we checked the antibody profile of S1-IgA after vaccination in the target groups and found that the titer gradually increased over

time in the convalescent group but remained significantly low in uninfected subjects (Figure 3 and Supplementary Table 3). On the other hand, the S1-IgM profile showed an insignificant difference between these two groups.

The global demand for the COVID-19 vaccine poses tremendous pressure on the manufacturer and supply chain. In this regard, the decision to administer a single dose of vaccine in the convalescent ones will spare another dose for a naïve person for a certain period [47]. With the increasing number of SARS-CoV-2 variants in the different region also raise question on the effectiveness of the vaccines in the long run [48]. Again, the length of SARS-CoV-2 vaccine-induced immunity in our

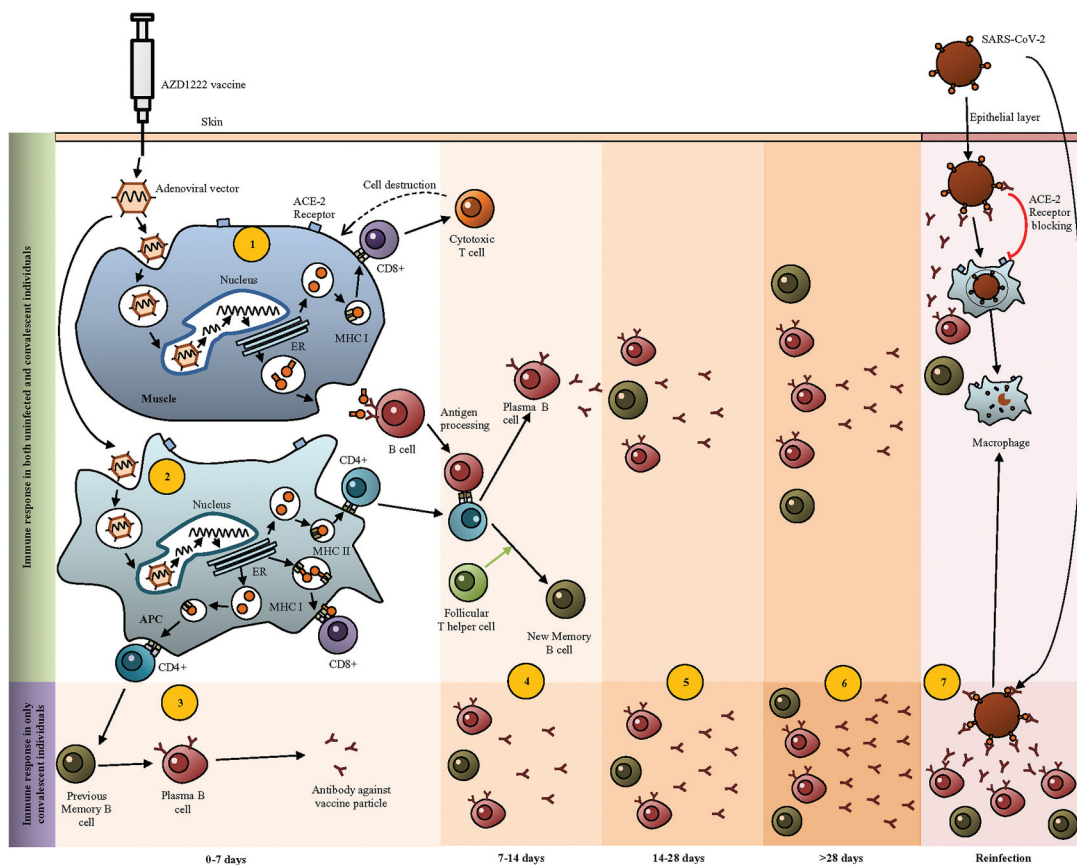


Figure 5. Probable mechanism of antibody production upon AZD1222 vaccination and reinfection. (1) Upon receiving the vaccine through the parenteral route, the adenovirus infects the muscle cells, expressing the viral antigens through the major histocompatibility complex class I (MHC I) molecule. The antigen-MHC I complex is identified by the CD8+ cells such as cytotoxic T lymphocyte (CTL) and helper T cells type 1 (Th1 cells), which activate CTL. This cascade leads to the release of cytokines which ultimately leads to the death of the infected cell. (2) The adenoviral particles are also endocytosed by the antigen-presenting cells (APCs), which then express antigens on its MHC class II (MHC II) complex. Upon expression of viral antigen on MHC II, complex CD4+ cells such as helper T cells (Th2 cells) are activated, activating B lymphocytes (B cells) into plasma B cells and memory B cells and memory Th2 cells. The plasma B cells produce antibodies against various antigenic components of the virus, essentially the spike protein. (3) If the individual has had previous exposure to the virus, then the memory Th2 cells and memory B cells are activated into both plasma and memory B cells. (4), (5), (6) With each passing day slowly but steadily, the amount of plasma B cells, memory cells, as well as antibodies increase in quantity for both previously infected and newly exposed individual, though at a much faster rate in previously infected individuals. (7) Upon reinfection, the memory Th2 cells and memory B cells formed due to the previous infection, and vaccination takes center stage and produces antibodies rapidly, minimizing the effect of the infection.

body is still not known. Therefore, countries should deploy strategies to combat the disease with long-term vision.

In a densely populated country like Bangladesh with over 160 million people, achieving herd immunity through vaccination is challenging. Recently, variants of different SARS-CoV-2 have been reported from Bangladesh, and cases of reinfection have become quite common among the general population and repeated reinfection among healthcare professionals [25,49,50]. In this context, how the immune system responds to a different variant of SARS-CoV-2 and what antibody level is needed in the system to arrest the virus without any symptoms would be remarkably interesting to know. The initial supply of the India-made AZD1222 vaccine in Bangladesh has already been administered, and the country is now looking for alternative sources. To get maximum protection against SARS-CoV-2, countries might need to administer two different vaccines covering mutations of concern of the spike protein upon careful consideration. Individuals who have received the first dose of AZD1222 may receive an inactivated virus as the second dose, which may mimic natural infection, thereby generating a robust immune response against many

viruses. However, follow-up studies will be required to measure the duration of the immune response after vaccination and the optimum interval between two to achieve maximum protection.

4.1. Limitations of the study

This study was conducted with a small group of subjects to observe the initial effect of the AZD1222 vaccine on the Bangladeshi population. Moreover, the impact of the first dose was only observed here. For longitudinal antibody dynamics study, more volunteers would be enrolled in future endeavors. Although S1 protein capable of binding neutralizing antibodies was used in this study, an appropriate neutralizing antibody test could significantly improve the quality of this work. Nevertheless, due to insufficient funds, a neutralizing assay or comparison of the results with commercially approved kits could not be performed. Furthermore, we were not able to study the role of cell-mediated immunity in relation to vaccination. Unfortunately, the questionnaire prepared to conduct the study didn't include Body Mass

Index (BMI) as a variable and thus couldn't collect such data from the subjects.

5. Conclusion

Our study presents that upon receiving the first dose of AZD1222, there was a rapid increase in the antibody levels of individuals who have been previously infected with SARS-CoV-2. This increase translates to more extended protection from SARS-CoV-2 and hopefully its variants. Concurrently, our study provides preliminary understanding for the combination of different types of vaccine, essentially, the combination of inactivated vaccine along with AZD1222.

Additionally, the study also represents that to optimize the distribution and allocation of the vaccine in the most efficient way, it would require the implementation of antibody tests simultaneously, essentially in resource strained countries. This would ensure that those who do not have antibodies be provided the first dose of the vaccination regarding those previously infected and represent higher antibody titer. This study group will investigate and monitor COVID-19 antibody dynamics after the second and future booster vaccination doses.

5.1. Recommendations

- (1) Antibody profiling before vaccination can be implemented because of the global scarcity of vaccine supply.
- (2) Subjects with recent RT-qPCR history and seroconverted ones can be enrolled for the latter phase in the vaccination program or administered a single dose of vaccine.
- (3) A person without prior COVID-19 history and seronegative for SARS-CoV-2 antibodies can be enrolled in the priority list, and two doses of vaccines can be administered.
- (4) Mix and match of vaccines can be implemented after proper approval from World Health Organization as soon as possible.

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