Original Article

Effect of Comorbidities on Antibody Status Following COVID-19 Vaccination – A Comparison between SARS-Cov-2 Infected and Non-infected Healthcare Professionalsin Dhaka, Bangladesh

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Abstract

Background: Vaccination with the Oxford-Astra Zeneca COVID-19 vaccine was initially started in the UK and quickly implemented across the globe including Bangladesh. ***Objective:*** To observe the difference in antibody status between infected and non-infected individuals as well as between relatively healthy individuals and individuals having comorbidities. ***Methods:*** This cross-sectional, analytical study was conducted in the Department of Biochemistry and Molecular Biology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh, between March 2021 and February 2022. A total of 70 adult participants (healthcare professionals) were included in this study who were working in different departments of BSMMU Hospital. Study participants were categorized into two groups: healthcare professionals who were infected by SARS CoV-2 and later vaccinated by two doses of AstraZeneca COVID-19 vaccine were included in group A, while group B included those who were not infected by SARS CoV-2 but took two doses of AstraZeneca COVID-19 vaccine. Each group had 35 participants.Demographic profile, detailed history was recorded in data collection sheet. Then blood pressurewas measured and recorded. Random blood sugar was estimated by glucose oxidasemethod, while serum IgG was assessed by chemiluminescent microparticle immunoassay method. ***Results:***Participants with hypertension in group A had IgG levels as median 2183.20 AU/ml, and IQR (inter quartile range)of 0 AU/ml, and in group B, as median 624.70 AU/ml, and IQR of 0 AU/ml. (P>0.05). In contrast, among participants with no hypertension showed significant differences in IgG levels (group A median 2242.65 AU/ml, and IQR 3758.88 AU/ml; group B median 619.60 AU/ml, and IQR 672.23 AU/ml) (P<0.001). Participants having both diabetes mellitus and hypertension in group A had IgG levels as median 1949.70 AU/ml, and IQR of 4294.43 AU/ml, and in group B, as median 739.00 AU/ml, and IQR of 423.75 AU/ml. (P<0.001). Among participants with no such comorbidities also showed significant differences in IgG levels (group A median 2183.20 AU/ml, and IQR 3547.50 AU/ml; group B median 592.40 AU/ml, and IQR 740.98 AU/ml) (P<0.001). After summation, participants having all types of comorbidities in group A had IgG levels as median 2183.20 AU/ml, and IQR of 4095.70 AU/ml, and in group B, as median 624.70 AU/ml, and IQR of 558.80 AU/ml. (P<0.001). In contrast, among participants with no comorbidities showed similar differences in IgG levels (group A median 2394.45 AU/ml, and IQR 3450.73 AU/ml; group B median 653.10 AU/ml, and IQR 990.13 AU/ml) (P<0.001).***Conclusion:***Antibody status (serum IgG levels) was significantly higher in previously infected vaccinated group (both with comorbidities and without comorbidities)than that of non-infected vaccinated group.

Keywords: COVID-19 Vaccination, antibody status, SARS CoV-2 infection, comorbidities, healthcare professionals

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Introduction

The SARS-CoV-2 pandemic has caused an unprecedented worldwidepublic health challenge.[1] In Bangladesh, coronavirus case was first reported on March 8, 2020, while the first death announced on March 18, 2020.[2]IgG antibody is associated with reduced risk of SARS-CoV-2 reinfection in the ensuring almost 6 months.[3] Scientists are trying their best to invent effective drug against COVID-19; however,none has come to the effect to date.Under the circumstances, the only way to protect the human being from the curse of COVID-19 by producing antibody either by low level passive exposure or active exposure to SARS-CoV-2 infection or vaccination or both.[4]Bangladesh started its nationwide administration of COVID-19 vaccine on February 7, 2021 with Oxford AstraZeneca produced and distributed by the Serum Institute of India.[5]Evidence showed a strong relationship between previous SARS-CoV-2 infection and higher antibody responses; individuals with previous SARS-CoV-2 infection generate strong humoral and cellular responses to one dose/two doses of COVID-19 vaccine, with evidence of high titres of in-vitro live virus neutralisation.[3] However, to date, no reports are available in our countryon the antibody status with or without previous SARS-CoV-2 infection and following as well as the antibody status of patients suffering with comorbidities like diabetes and hypertension. Moreover, it is unknown how much antibody level raised after SARS-Cov-2 infection and following vaccination, and whether that level is enough to protect the human being from reinfection or hospitalization.[6]Evidence showed that patients with type 2 and type 1 diabetes or cardiovascular diseases (CVD) have an increased vulnerability to severe sufferings from SARS-CoV-2.[1,4] Therefore, vaccination should be prioritized in diabetes, hypertensive,and CVD patients.Moreover, we also felt the necessity to know the diversity in immunity status ofpeople in different healthcare settings, as they remain most vulnerable in this pandemic situation. Hence, we proposed this cross-sectional, analytical study to evaluate and compare the antibody status between SARS-CoV-2 infected vaccinated and SARS-CoV-2 non-infected vaccinated healthcare professionals, as well as observe the difference in antibody status between subjects having comorbidities and without any comorbidity.

Methods

This cross-sectional, analytical study was conducted in the Department of Biochemistry and Molecular Biology, Bangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh, between March 2021 and February 2022.

Inclusion criteria:

1) Aged between 25 and 65 years;
2) Healthcare professionals who were SARS-CoV-2 infected last 8-12 months ago (RT-PCR positive report) and received two doses of AstraZeneca COVID-19 vaccine 4 to 6 months back; and
3) Healthcare professionals who were SARS-CoV-2 non infected but received two doses ofAstraZeneca vaccine last 4 to 6 months ago.

Exclusion criteria:

1) Subject with acute infection;
2) Pregnant women;
3) Lactating mother;
4) History of heart failure;
5) Chronic systemic diseases,e.g., chronic liver disease, chronic kidney disease; and
6) Subject who are suffering from any immunosuppressive disorderse.g., cancer, SLE, etc.

Based on inclusion and exclusion criteria, a total of 70 healthcare professionals were included in this study from different departments of BSMMU Hospital. Study participants were categorized into two groups: group A consisted of healthcare professionals who were previously infected by SARS CoV-2 and later vaccinated (two doses of AstraZeneca COVID-19 vaccine) and group B by who were not infected by SARS CoV-2 but received the same doses of AstraZeneca COVID-19 vaccine. There were 35 participants in each group. A data collection sheet formatted both in English and Bengali was used as a data collection tool. The sheet included three sections: section-I contained general information, while section-II contained information related to SARS-CoV-2 infection and section-III included further test reports related to this study.

Demographic profile, detailed history was recorded

in data collection sheet.Then bloodpressure of each individualwas measured and recorded. After that, with all aseptic precaution, 5ml blood sample was collected from the anti-cubital vein, using a disposable plastic syringe. 2ml of blood was delivered immediately into sodium-fluoride tube (grey top tube) and 3ml into a plain tube (red top tube). All the test tubes were centrifuged properly at 3000 rpm for 10 minutes to separate plasma and serum within one hour of collection. Then the serum (about 500µml) was separated from each of the plain tube by micropipette, collected in Eppendorf tube, properly labeled, and stored at minus 65-degreeCelsius temperature. Separated plasma was used for estimation of random blood sugar (RBS) by using by glucose oxidase method. Estimation of serum IgG levels wasdoneusing chemiluminescent microparticle immunoassay in Abbott AlinityiAutoanalyzer (made by Abbott Inc.,USA). All the biochemical and immunological assays were performedin the Department of Biochemistry and Molecular Biology ofBangabandhu Sheikh Mujib Medical University (BSMMU).Autoanalyzer used in this study was calibrated before starting the tests as per test manual. Before starting daily investigations, control run was done. Quality control and quality assurance in all areas were maintained as per respective laboratory rules. Pre-analytic, analytic, and post-analytic errors were carefully minimized as per laboratory standard operating procedure (SOP).

After multiple checking, data were recorded in a predesigned data collection sheet. Continuous variables were expressed as mean±SD and compared between groups by unpaired student's t-test. Categorical variables were expressed as frequency and percentage and compared using Chi-square test. Mann-Whitney U test was done to compare serum IgG levels in between SARS-CoV-2 infected vaccinated group and SARS-CoV-2 non-infected vaccinated group.Level of significance was defined as P value <0.05 at 95% confidence interval. Statistical analysis was done using Statistical Package for the Social Sciences (SPSS) version 20.0 for windows.

Results

The mean age of previously infected and vaccinated individuals (group A) was 41.14±12.51 years, while 38.43±9.18 years for non-infected but vaccinated individuals (group B). However, there was no significant difference in age between the groups (P>0.05). A male predominance was observed in group A; in contrast, female predominance was found in group B. The difference in gender between the groups was statistically significant (P<0.05). In group A, there were 57.1% doctors, 20% nurses, 8.6% phlebotomists, and 14.3% other staff. Similarly, in group B, there were 48.6% doctors, 17.1% nurses, 11.4% phlebotomists, and 22.9% other staff. No significant difference was observed between the groups (P>0.05) (Table 1).Mean systolic blood pressurein group A was 121.00±7.05mm of Hg, while in group B 119.37±7.92 mm of Hg. Observed mean diastolic blood pressure were 80.14±6.36 mm of Hg and 79.43±5.53 mm of Hg respectively. Random blood sugar was found 6.05±1.97 mmol/L in group A, whereas 5.91±1.88 mmol/L in group B. However, the differencesbetween the groups were not statistically significant in any of those parameters (P>0.05) (Table 2). Participants withhypertension had following IgG levels: in group A as median 2183.20 AU/ml, and IQR of 0 AU/ml, and in group B, as median 624.70 AU/ml, and IQR of 0 AU/ml. (P>0.05). In contrast, among participants with no hypertension showed significant differences in IgG levels (group A median 2242.65AU/ml, and IQR 3758.88AU/ml; group B median 619.60AU/ml, and IQR 672.23AU/ml) (P<0.001) (Table 3). Participants having both diabetes mellitus and hypertension had following IgG levels: in group A as median 1949.70AU/ml, and IQR of 4294.43 AU/ml, and in group B, as median 739.00AU/ml, and IQR of 423.75AU/ml. (P<0.001). Among participants with no such comorbidities also showed significant differences in IgG levels (group A median 2183.20AU/ml, and IQR 3547.50AU/ml; group B median 592.40AU/ml, and IQR 740.98AU/ml) (P<0.001) (Table 4). After summation, participants having all types of comorbidities had following IgG levels: in group A as median 2183.20 AU/ml, and IQR of 4095.70 AU/ml, and in group B, as median 624.70 AU/ml, and IQR of 558.80 AU/ml. (P<0.001). Incontrast, among participants with no comorbidities showed similar differences in IgG levels (group A median 2394.45 AU/ml, and IQR 3450.73 AU/ml; group B median 653.10 AU/ml, and IQR 990.13 AU/ml) (P<0.001) (Table 5).

Table 1. Demographic characteristics of the study participants (n=70)

Variables	Group A (n=35)	Group B (n=35)	P value
Age in years			
Mean±SD	41.14±12.51	38.43±9.18	>0.05^{NS}
Gender			
Male	24 (68.6)	14 (40.0)	<0.05^{S}
Female	11 (31.4)	21 (60.0)	
Occupation			
Doctor	20 (57.1)	17 (48.6)	>0.05^{NS}
Nurse	7 (20.0)	6 (17.1)	
Phlebotomist	3 (8.6)	4 (11.4)	
Other Staff	5 (14.3)	8 (22.9)	

Continuous variables were expressed as mean±SD, while categorical variables were expressed as frequency and percentage. Unpaired students t-test was used to compare differences in age, while Chi-square test was used to compare gender and occupation. S=significant, NS=not significant.

Table 2. Clinical characteristics of the study participants (n=70)

Variables	Group A (n=35)	Group B (n=35)	P value
Systolic blood pressure mm of Hg	121.00±7.05	119.37±7.92	>0.05^{NS}
Diastolic blood pressure mm of Hg	80.14±6.36	79.43±5.53	>0.05^{NS}
Random blood sugar mmol/L	6.05±1.97	5.91±1.88	>0.05^{NS}

Data were expressed as mean±SD. P value reached from Chi-square test; NS=not significant.

Table 3. Antibody status of the study subjects with or without hypertension (n=70)

Hyper-tension	Antibody status (AU/mL)	Group A (n=35)	Group B (n=35)	P value
Present	Median	2183.20	624.70	>0.05^{NS}
	IQR	0.00	0.00	
	Min-max	1036.20 -5131.90	259.80 -764.50	
Absent	Median	2242.65	619.60	<0.001^{S}
	IQR	3758.88	672.23	
	Min-max	861.70 -12884.10	96.10 -2330.00	

Data were expressed as median and IQR (interquartilerange).P value reached from Mann-Whitney U test; NS=not significant, S=significant.

Table 4. Antibody status of the study subjects with or without diabetes mellitus and hypertension (n=70)

Both diabetes mellitus &hypertension	Antibody status (AU/mL)	Infected vaccinated	Non infected vaccinated	P value
Present	Median	1949.70	739.00	<0.05^{S}
	IQR	4294.43	423.75	
	Min - max	897.3-8797.6	232.3-918.2	
Absent	Median	2183.20	592.40	<0.001^{S}
	IQR	3547.50	740.98	
	Min - max	861.7-12884.1	96.1-2330.0	

Data were expressed as median and IQR (interquartile range).P value reached from Mann-Whitney U test; S=significant.

Table5. Antibody status of the study subjects with and without comorbidities (n=70)

Variables	Antibody status (AU/mL)	Group A	Group B	P value
With comorbidity (n=26)	Median	2183.20	624.70	<0.001^{S}
	IQR	4095.70	558.80	
	Min - max	897.30-8797.60	99.40-1393.90	
Without comorbidity (n=44)	Median	2394.45	653.10	<0.001^{S}
	IQR	3450.73	990.13	
	Min - max	861.70-12884.10	96.10-2330.00	

Data were expressed as median and IQR (interquartile range). P value reached from Mann-Whitney U test; S=significant.

Discussion

Antibody plays a vital role in suppressing the pathogenesis of SARS-CoV-2 by disrupting the binding of viral spike protein to angiotensin-conveting-enzyme2 receptor on the target cell.[6]A longitudinal study in China showed that IgM levels increased first week after SARS-CoV-2 infection peaked 2 weeks after that decline whereas IgG was detectable after 1 week and maintained at a high level for a long period.[7]The peripheral T and B cell from the SARS-CoV-2 patients revealed a positive correlation of humoral immune response and the T cell immune memory with disease severity.[8]

Evidence showed a strong relationship between previous SARS-CoV-2 infection and higher

antibody responses. Several research reported that individuals with previous SARS-CoV-2 infection generate strong humoral and cellular responses to one dose/two doses of COVID-19 vaccine, with evidence of high titres of in-vitro live virus neutralisation. In contrast, most individuals who are infection-naive generate both weak T-cell responses and low titres of neutralising antibodies.[9-14] Our results are in congruence with those research fundings.

In our study, it was observed that in both group a considerable number of study subjects were suffering from hypertension, or diabetes or both. We found that 22.9% infected participants had some types of comorbidities. Yang et al.[15]reported that prevalence of SARS-CoV-2 was higher with hypertension 21.1% and diabetes 9.7%. Similarly, Sanyaolu et al.[16]found that most common comorbidities of SARS-CoV-2 patients were hypertension (15.8%) and diabetes (9.4%).Studies also found that the relative risk of developing severe COVID-19 or death is higher in patients with risk factors for CVD (hypertension, diabetes) and much higher in patients with CVD.[1,4,17,18] Similarly, a study conducted in Bangladesh found that COVID-19 patients with CVD had almost five times higher odds of death, and COVID-19 patients with CVD and diabetes had almost seven times higher odds of death.[19]

We evaluated the antibody level between hypertensive and non-hypertensive individuals in between infected vaccinated group and non-infected vaccinated group; we found a higher antibody level in infected vaccinated group than that of non-infected vaccinated group. Simultaneously, wealso evaluated the antibody level with or without both diabetic and hypertensive individuals in infected vaccinated and non-infected vaccinated group; higher antibody levels in infected vaccinated group compared to non-infected vaccinated group was also observed. Ali et al.[20]stated that diabetic and hypertensive individuals had a robust antibodyresponse to vaccination as demonstrated by their high antibody titer which was statistically significant. In their study, done in Kuwait, three weeks after second dose of vaccine they observed that serum IgG level was 138 BAU/ml in diabetic participants and without diabetic participants was 154 BAU/ml, while in hypertensive individuals 144 BAU/ml and non-hypertensive individuals 151 BAU/ml, which were relatively lower.[20] Another study done in Austria bySourij et al.[21] reported that after the first vaccination only 52.7% type-1diabetes group and 48.0% in the type-2 diabetes group showed antibody level above the cut-off value but the antibody level after the second vaccination were similar in type-1, type-2 and healthy controls. Another study done by Iacobucci et al.[22]suggested that after a single dose of vaccination there was significant difference in antibody level in between diabetes, cardiovascular disease and normal individuals; however, in other study done by Uysal et al.[23] observed that the inequalities in antibody levels amongst those groups did not persist after the second dose, as high antibody titers>250 U/ml were observed nearly all participants. Our findings are more or less similar to those studies.

Conclusion

To summarize, antibody status (serum IgG levels) was significantly higher in previously infected vaccinated group (both with comorbidities and without comorbidities) than that of non-infected vaccinated group. However, further studies are recommended involving larger samples from different age groups and multicentre across the country.

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Conflict of interest: The authors declare no conflict of interest.

Ethical approval: The study was approved by the Institutional Review Board ofBangabandhu Sheikh Mujib Medical University (BSMMU), Dhaka, Bangladesh.

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Authors' contribution: Concept and design of the study: RR, FHM; Subject selection and collection of samples: RR, FHM, MM, SA, TC, FS; Data collection and compilation: RR, FHM; Data analysis: RR, MM; Manuscript writing, revision and finalizing: RR, FHM, MM, SA, TC, FS.

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