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Efficacy of Two Vaccine Platforms against SARS-CoV-2

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ABSTRACT

Introduction: Several vaccine platforms have been designed to elicit an effective immune response against SARS-CoV-2. This study was aimed to evaluate the humoral immune response in Iranian people vaccinated with both inactivated virus vaccines and vector vaccine platforms. **Methods:** The study enrolled 360 vaccinated individuals with inactivated virus vaccines (BBIBP-CorV and Covaxin) and vector vaccine platforms (AstraZeneca and Sputnik V). Serum samples were collected for each volunteer on days 14 to 21 after vaccination, and anti–SARS-CoV-2 spike receptor-binding domain (RBD) IgG concentrations were analyzed by ELISA. **Results:** Higher antibody titers were observed in participants vaccinated with vector vaccines compared with those immunized with inactivated virus, especially in subjects who received two doses of AstraZeneca. (AstraZeneca: 204.19 U/mL [95% CI, 175.5-232.2] vs. Sputnik V :114.67 U/mL [95% CI, 99.54-129.8]; $P = 0.007$). It was also observed that antibody titers were not significantly different between the two groups receiving inactivated vaccines ($P = 0.86$). Our results indicated that 28% of the population vaccinated with Covaxin and 32% of people vaccinated with BBIBP-CorV showed no response to the vaccine. There was also a statistically insignificant difference between age, BMI, and gender with antibody level in each group of vaccines ($P > 0.05$). **Conclusion:** Based on a limited population data , our study showed that the viral vector-based vaccines produced higher levels of neutralized antibodies than the inactivated vaccines, and their rate of non-response was lesser.

INTRODUCTION

The worldwide pandemic of Coronavirus disease 2019 (COVID-19) was originated in Wuhan, China. COVID-19 is a novel emerging infectious disease caused by a new type of coronavirus, namely, SARS-CoV-2. Effective vaccines are urgently needed due to the 2% mortality rate and profound medical, economic, and social implications of this pandemic [1]. The SARS-Cov-2 virus has four protein structures: Spike protein, Envelope protein, Membrane protein, and Nucleo-capsid protein. The virus binds to specific ACE2 receptors on the cell surface via the S protein (Spike) and infects the cells. Therefore, neutralizing antibodies against the S protein can inhibit this process and prevent the virus invasion. Accordingly, most vaccines developed for SARS-CoV-2 are designed to produce antibodies against the SARS-CoV-2's spike protein [2].

SARS-CoV-2 vaccines are one of the most important public health measures to prevent and reduce the morbidity and mortality of SARS-CoV-2 infection. Various vaccine candidates have been developed to generate protective immune responses against the spike antigen of SARS-CoV-2 [3].

There are several platforms of SARS-CoV-2 vaccines that include inactivated virus vaccines, vector vaccines, DNA vaccines, and mRNA vaccines. Each different type of the vaccines has its own advantages and disadvantages [4]. Authorized vaccines prevent COVID-19 by inducing the production of antibodies against a specific virus protein.

The Sputnik V vaccine contains adenovirus DNA, a recombinant adenovirus type 26 (rAd26) vector, and a recombinant adenovirus type 5 (rAd5) vector. Both vectors are used as containers to deliver SARS-CoV-2's spike glycoprotein (i.e., rAd26-S and rAd5-S) to cells and synthesize new coronavirus's envelope proteins. The full-length SARS-CoV-2 glycoprotein S is integrated into the vector and heterogeneous boosting with 2 different vectors for 2 vaccine shots is planned. In this manner, a more sustainable immunity is produced than vaccines which use the exact delivery mechanism for the both

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shots. The phase 3 clinical trial showed this vaccine-induced robust humoral and cellular immune responses in all age groups [5]. The administration route is intramuscular with 0.5 mL/dose, and there is a 21-day interval between the first dose (rAd26) and the second dose (rAd5) [6, 5].

The ChAdOx1 nCoV-19 vaccine (AZD1222) was developed at Oxford University, U.K., and AstraZeneca. It was authorized for emergency use in the U.K. on Dec 30, 2020, based on a regimen of two standard doses (0.5 ml/dose) given with an interval of 4 –12 weeks for adults aged 18 years and older. The ChAdOx1 nCoV-19 vaccine consists of a replication-deficient chimpanzee adenoviral vector ChAdOx1, containing a full-length SARS-CoV-2 spike protein (nCoV-19) gene [7]. Clinical trials have shown that ChAdOx1 nCoV-19 vaccine induces neutralizing antibodies and CD4+ and CD8+ T cell subsets against the SARS-CoV-2's spike protein [8, 9].

The Beijing Institute of Biological Products generated BBIBP-CorV vaccine as an inactivated vaccine in early 2020. The BBIBP-CorV mechanism induces the immune system to make antibodies against SARS-CoV-2, inducing robust humoral responses and memory T cells and B cells. These antibodies then recognize and attach to the viral proteins, such as the spike proteins that appear on the virus surface. Inactivated viruses are used in BBIBP-CorV vaccine, including three inactivated coronavirus variants by beta-propiolactone. Inactivated viruses could no longer replicate while the antigenic properties of their proteins, including the spikes, remains intact. The inactivated viruses mixed with a tiny amount of an adjuvant based on aluminum could help to stimulate the immune system to boost its response against the infection. Inactivated SARS-CoV-2 is administrated in 0.5 mL/dose by an intramuscular injection of two doses (one dose/21-28 days) [10, 11].

Covaxin (BBV152) is a whole-virion inactivated SARS-CoV-2 vaccine developed by India-based Bharat Biotech and the Indian Council of Medical Research. The vaccine was developed by β-propiolactone inactivation of 3 μg or 6 μg SARS-CoV-2 strain (Strain: NIV-2020-770) and formulated with a toll-like receptor 7/8 agonist molecule (IMDG) adsorbed to alum (Algel). The Covaxin (BBV152) is a two-dose (0.5 ml/dose) vaccination regimen given 4 weeks apart. Covaxin induces both humoral and cell-mediated responses [12-14]. The aim of this study was to evaluate the humoral immune response in the serum of Iranian people vaccinated with both inactivated virus vaccines and vector vaccine platforms.

MATERIALS AND METHODS

Ethics Statement and Study Population and Design

The Ethics Committee of Arak University of Medical Sciences (Arak, Iran) approved this study protocol by IR.ARAKMU.REC.1400.024 authorization. This study was conducted on 360 people vaccinated with 4 different types of vaccines (90 people for each vaccine). All volunteers were selected among the hospital and Arak Medical University staff and tested for anti- SARS-CoV-2 antibodies after the vaccinations. Before starting the study, informed consent from the patients who participated in the research were obtained. Participants in the study completed a questionnaire that included information on age, gender, weight, height, type of vaccine, date of administration of vaccine dose, and adverse effects. The inclusion and exclusion criteria of the study are listed in Table 1.

Inclusion Criteria	Males and females within the age range from 20 to 60 years
	Written informed consent
	Subject body mass index (BMI): 18 ≤ BMI ≤ 30
	Absence of COVID-19 diagnosis in medical history
	Absence of acute infectious diseases at the time of vaccine administration and 14 days before vaccination
	The absence of malignant diseases
Exclusion Criteria	Any vaccination over the last 30 days
	History of COVID-2019 disease
	Positive PCR test results for SARS-CoV-2
	Positive test results for IgM and IgG antibodies to SARS-CoV-2
	Respiratory symptoms in the last 14 days
	The administration of immunoglobulins or other blood products in the last 3 months
	History of chronic alcohol abuse and/or drug use
	The subject has received immunosuppressive and/or immunomodulating agents within 6 months before the start of the study
	Pregnancy or breastfeeding
	A history of autoimmune diseases in the volunteer's medical history
	Simultaneous treatment with immunosuppressive drugs, incl. corticosteroids (2 weeks) 4 weeks before study drug administration
	Subjects who received antiviral drugs, immunoglobulins or blood transfusions, or any other investigational drug within 4 weeks before the study drug administration
	Subjects who received anti-inflammatory drugs 2 days before the study

Table 1. Study inclusion and exclusion criteria.

Laboratory Analyses

Serum samples were collected for each volunteer on days 14 to 21 after the vaccination, and virus-neutralizing antibodies were measured. Also, measurable hematological indicators were evaluated during the CBC test of these people. SARS-CoV-2 antibody levels were measured by ELISA detecting anti-S1-RBD IgG (DiaZist Cat No: DG. COVS.01). Normalized results were reported in A.U./ml. In this method, the assayed antibodies were captured by the recombinant SARS-CoV-2 stuck to the bottom of wells and anti-human IgG conjugated with HRP. According to the kit's instruction, the assay was performed using a threshold of 11 A.U./ml to identify positive samples. The lower amount of 9 A.U./ml was considered as a negative sample. Results were expressed as optical density (OD_{450}) measurements. According to the manufacturer's instructions, the concentration of each sample was achieved by calculating the standard curve. The cutoff for seropositivity was set at 0.76, and all samples below these thresholds were considered negative.

Statistical Analysis

The volunteers were divided into 4 groups according to their vaccinations. The mean, standard deviation, and *P* values were analyzed for each group. One-way ANOVA ($P < 0.05$) followed by Tukey's post-hoc statistical analysis was used for

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multiple comparisons. Statistical analyses were performed using the IBM SPSS Statistics software.

Characteristics	Sputnik V	AstraZeneca	BBIBP-CorV	Covaxin	Sig
Gender					
Female	47	46	45	44	-
Male	43	44	45	46	-
Age					
Mean	42.6981	37.8846	43.4516	39.3929	0.82
Std. Deviation	9.30795	10.97172	11.12606	8.41665	0.74
BMI					
Mean	24.7519	24.4637	25.2620	25.4779	0.719
Std. Deviation	3.79938	4.88005	4.37997	4.28856	

Table 2. Demographic characteristics in COVID-19 vaccine recipients.

In these 360 subjects, anti-S-RBD IgG levels for each type of the vaccine were not significantly different between women and men ($P > 0.05$; Fig. 1). Additionally, median anti-S1 titers were independent of BMI, and there was no significant difference between BMI and antibody levels in each group of the vaccines ($P > 0.05$; Table 2).

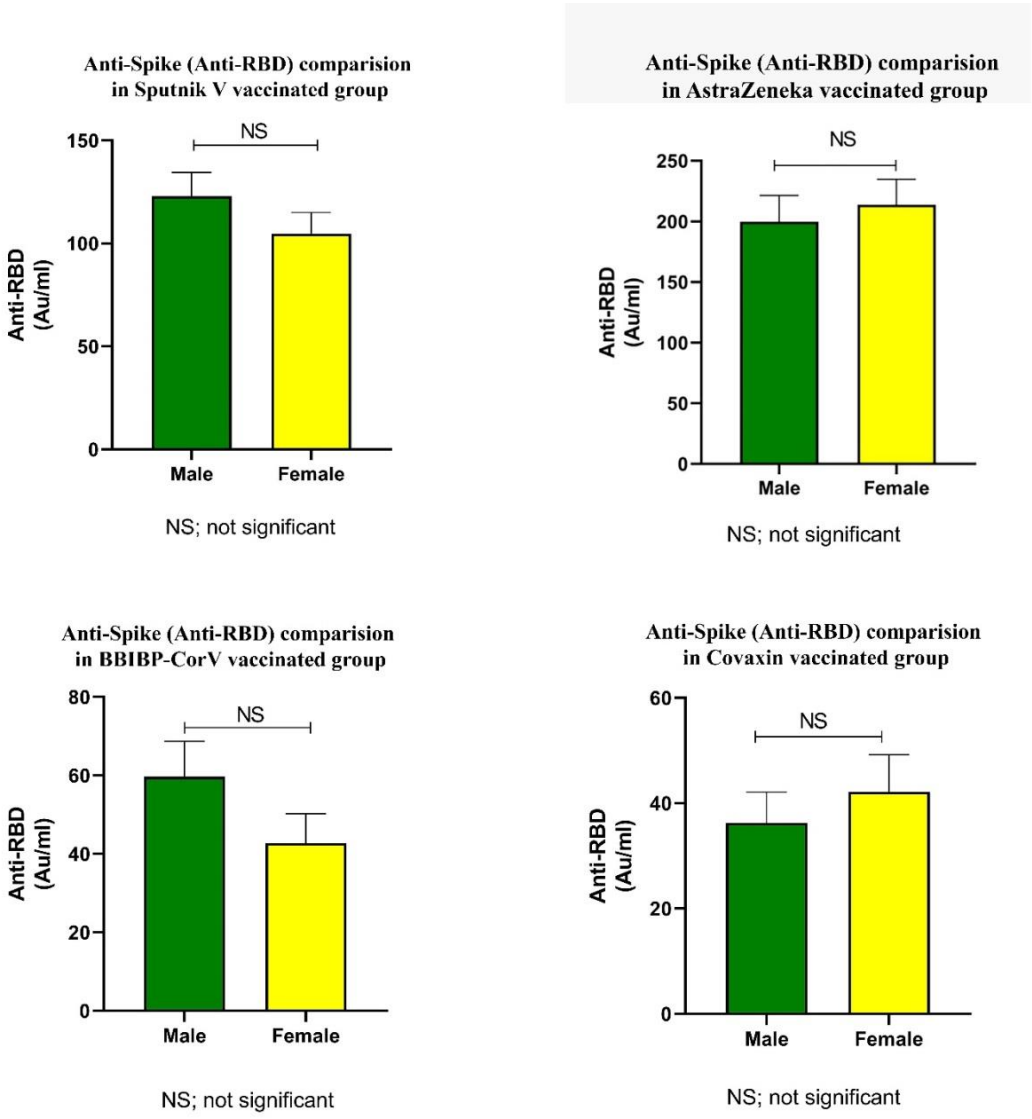

Fig. 1. Comparison of the antibody titer with gender. There was no significant difference between women and men with respect to their antibody levels in each vaccine group; Data points show mean ± S.D.; the error bars reflect S.D. NS: not significant.

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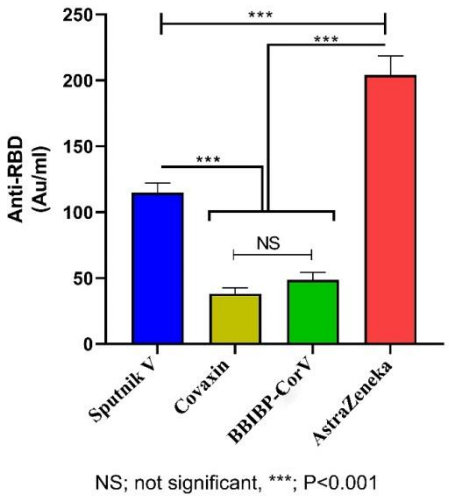
The information on the adverse effects of vaccination was evaluated in each vaccinated group. About 77.2 % of the subjects displayed one or more symptoms after the vaccination. After the second dose, the highest adverse effects were shown in subjects vaccinated with AstraZeneca (86 %), and the lowest adverse effects were detected in subjects immunized with BBIBP-CorV (50 %). Differential blood cell counts were not significantly different among subjects who received the COVID-19 vaccines ($P > 0.05$; Table 3).

Table 3. Comparison of hematologic features in COVID-19 vaccine recipients.

Characteristics	Sputnik V	AstraZeneca	BBIBP-CorV	Covaxin	Sig.
WBC					
Mean	7.0314	6.6609	6.7238	6.9091	0.8
Std. Deviation	1.72401	2.04911	1.34978	1.34267	
PLT					
Mean	252.0600	232.1739	226.7143	233.5909	0.215
Std. Deviation	58.00331	50.24725	48.39436	53.67402	
Lymphocyte					
Mean	34.6314	36.1217	36.0762	34.1045	0.579
Std. Deviation	6.91160	5.52366	4.83641	6.46393	
Neutrophil					
Mean	56.3432	53.3739	54.9952	57.7333	0.202
Std. Deviation	7.75492	7.77737	4.97629	6.90234	

Comparison of Antibody Titers in the Studied Groups

As indicated in Fig. 2, members of two groups who were vaccinated with the vector vaccines had higher antibody titers than those immunized with the inactivated virus vaccines. Higher antibody titers were observed in subjects vaccinated with 2 doses of AstraZeneca (204.19 U/mL; 95% CI, 175.5-232.2), compared to Sputnik V recipients (114.67 U/mL; 95% CI, 99.54-129.8); $P = 0.007$. Antibody titers were not significantly different between the two groups who had received Covaxin and BBIBP-CorV inactivated vaccines ($P = 0.86$).



NS; not significant, ***; P<0.001

Fig. 2. Comparison of anti-Spike IgG (anti-RBD) in different vaccinated groups.

DISCUSSION

Vaccines are effective to control the spread of SARS-CoV-2 pandemic among the population [15]. To monitor the immunity induced by a vaccine, it is necessary to measure the level of antigen-specific antibodies to evaluate its effectiveness [16]. In this study, we measured the level of anti-S-RBD IgG, following vaccination with four different vaccine types against COVID-19 used in Iran, namely, Sputnik V, AstraZeneca, BBIBP-CorV, and Covaxin.

The main findings of the present study indicated that the level of antibodies was significantly higher in AstraZeneca recipients. In addition, among the vector-based vaccine group, individuals immunized with Sputnik V vaccine produced considerably lower levels of antibodies compared to people vaccinated with AstraZeneca. Considering that Sputnik V vaccine is based on Ad5 vector and previous studies have shown that the majority of the population (65 to 100% of Africans, 30 to 80% of Asians, 61% of Europeans, and 37 to 70% of Americans), have a high titer of pre-existed Ad5 antibodies due to their previous Ad5 infections, it can be assumed that high titers of anti-Ad5 antibodies can suppress the immunogenicity of Ad5 vector-based vaccines. This increase in anti-Ad5 antibody titers in populations, especially in Africa and Asia, has raised concerns about using

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Vaccine Research

Ad5-based vaccines [17, 18]. Therefore, the lower antibody titer in the Sputnik group compared to than the AstraZeneca group in our study, is possibly due to the presence of anti-Ad5 antibodies.

On the other hand, the results of the previous studies indicate that since the Oxford/AstraZeneca vaccine production relies on chimpanzee adenovirus, the concern about the presence of primary antibodies against the vector and the previous immunogenicity problem with these adenoviruses (except in some African regions) could partially be dismissed. Therefore, we reckon that the high anti-SARS-Cov-2 antibody titer in the AstraZeneca-vaccinated group could be due to the lesser exposure of the majority of the study population and, thus, the lack of previous immunity, to chimpanzee adenovirus [2].

Our results showed that 28% of the population vaccinated with Covaxin and 32% of people vaccinated with BBIBP-CorV had shown no response to the vaccine. The approved production technology of inactivated vaccines for BBIBP-CorV and Covaxin is similar. In this method, the virus is cultured in Vero cells and then is chemically inactivated by beta-propiolactone (BPL). Finally, after the purification, the proper adjuvant is added to the vaccine. Unlike the vector vaccines which maintain the native form of the spike protein, several studies performed on the molecular structure of the BPL-deactivated SARS-Cov-2 virus have shown that inactivated virus surface spikes form a post-fusion structure. In contrast, other data suggest that the spike protein of formalin-inactivated viruses show a pre-fusion structure [19-21]. Genetic modifications are done during the vaccine preparation to prevent the formation of post-fusion structure in the S protein; hence, the produced S protein would have a pre-fusion structure [22, 23]. Thus, participants in our study vaccinated with BBIBP-CorV and Covaxin vaccines may have high titers of binding but non-neutralizing antibodies, despite high levels of the neutralizing antibodies. These neutralizing antibodies can play an essential role in protection through practical Fc-mediated functions, including antibody-dependent phagocytosis, cytotoxicity, and activation of antibody-dependent natural killer cells [24].

Also, age, BMI and gender did not affect the immune response among all vaccinated groups. Our findings of the non-significant antibody titers in the vaccinated individuals, matched with age, were consistent with the phase 3 clinical trial of the Sputnik V vaccine. These data indicated that the antibody levels after the vaccination were unrelated to age [25]. This was consistent with another study in the U.K., where the evaluated antibody titers in vaccinated people with two dosages of the AstraZeneca vaccine were found to be similar in immunogenicity in all age groups [26]. However, in a study by Müller and colleagues, a lower titer of neutralizing antibodies was reported in the elderly group after vaccination with Biontech/Pfizer BNT162b2. This data indicates that this population needs precise monitoring and may require prior immunization and/or increased vaccine doses to ensure long-term safety and protection against the infection [27].

According to the questionnaires completed by the participants, among the four different types of used vaccines, the most side-effects were reported for the AstraZeneca group; however, these side-effects were moderate, and the lowest rate of side-effects was observed for the BBIBP-CorV group. Our finding was consistent with studies in the United Arab Emirates and India. A study in the United Arab Emirates showed that the side effects of the BBIBP-CorV vaccine were mild, and the majority of participants in the study had low and predictable side-effects while none experienced severe manifestations and required hospitalization. Although there is no satisfactory definition related to COVID-19 immunity, high titers of neutralizing antibodies and a strong CD8+ and CD4+ T-cells response play an essential role in developing protective immunity to the infection [8].

Although, the acquired immunity against SARS-CoV-2 is dependent on the presence of an antibody response, it would be impossible to determine the protective immunity threshold against COVID-19 by measuring the antibody titer alone. Functional assays that detect neutralizing antibodies are needed to understand better and define vaccination immunity, such as virus-neutralization assays [28]. Therefore, the lack of information on the neutralizing activity of the sera of the vaccinated people and the cellular response data could be considered as limitations of the present study. Also, the side-effects of the vaccines have not been formally evaluated. Other limitation of our study could be the small number of participants and not including older people with underlying diseases. Hence, larger groups are suggested to be included in the future studies to accurately measure the antibody response and compare the antibody titer across the age and gender. In conclusion, our study showed in a limited population in Iran, that viral vector-based vaccines produce higher levels of neutralized antibodies than inactivated vaccines, and their rates of non-response was lesser.

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CONFLICT OF INTERESTS

The authors declare they have no conflict of interests.

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