Evaluation of Antibody Response After COVID-19 Vaccination in Healthcare Workers: A Turkish Tertiary Hospital Experience

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ABSTRACT

Objective: It was aimed to explore the correlation between demographic characteristics and the presence of comorbidity in the antibody response after the second dose of vaccination in healthcare workers in this study. In addition, the third and fourth dose approaches of the participants and the protection of the antibody levels formed by the two doses of vaccine against COVID-19 were examined observationally.
Methods: Health workers, whose NCP and S protein antibody levels were detected on the 30th day after the second dose of the CoronaVac vaccine, were followed up in terms of being vaccinated for the third and fourth doses and having COVID-19.
Results: Higher levels of S antibodies were detected in women after two doses of vaccination (*p=0.001*). It was pointed out that smoking has a negative effect on the antibody response after vaccination (*p=0.008*). People who had pre-vaccine COVID-19 had higher NCP antibody levels after two doses of vaccination (*p=0.013*). Of the participants, 152 (97.4%) were vaccinated with the third dose and 110 (70.5%) with the fourth dose.
Conclusion: The antibody response after two doses of inactivated CoronaVac® vaccination is higher in women, young people, non-smokers and people previously infected with SARS-CoV-2.

Keywords: COVID-19 Vaccines, Vaccine, Antibody Response, Protection, Healthcare Workers

Sağlık Çalışanlarında COVID-19 Aşısı Sonrası Antikor Yanıtının Değerlendirilmesi: Türkiye'de Üçüncü Basamak Bir Hastane Deneyimi

ÖZET

Amaç: Bu çalışmada sağlık çalışanlarında ikinci doz aşılama sonrası antikor yanıtında komorbidite varlığı ile demografik özellikler arasındaki ilişkinin araştırılması amaçlanmıştır. Ayrıca katılımcıların üçüncü ve dördüncü doz yaklaşımları ile iki doz aşının COVID-19'a karşı oluşturduğu antikor düzeylerinin koruyuculuğu gözlemsel olarak incelenmiştir.
Yöntem: CoronaVac aşısının ikinci dozundan sonraki 30. günde NCP ve S protein antikor düzeyleri saptanan sağlık çalışanları, üçüncü ve dördüncü doz aşılarını yaptırmaları ve COVID-19'a sahip olmaları açısından takibe alınmıştır.
Bulgular: İki doz aşılamadan sonra kadınlarda daha yüksek S antikorları saptanmıştır (*p=0.001*). Sigaranın aşılama sonrası antikor cevabını olumsuz etkilediğine dikkat çekilmiştir (*p=0,008*). Aşı öncesi COVID-19 enfeksiyonu olan kişilerin, iki doz aşılamadan sonra daha yüksek NCP antikor seviyelerine sahipolduğu belirlendi (*p=0.013*). Katılımcıların 152'si (%97,4) üçüncü doz, 110'u (%70,5) dördüncü doz ile aşılanmıştır.
Sonuç: İki doz inaktif CoronaVac® aşılama sonrası antikor yanıtı, kadınlarda, gençlerde, sigara içmeyenlerde ve daha önce SARS-CoV-2 ile enfekte olmuş kişilerde daha yüksektir.

Anahtar Kelimeler: COVID-19 Aşıları, Aşı, Antikor Yanıtı, Koruma, Sağlık Çalışanları

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Introduction

The SARS-CoV-2 virus, which arose in December 2019 in China, quickly affected the whole world, causing the Coronavirus disease 2019 (COVID-19) pandemic. This pandemic has caused millions of deaths around the world, with portentous effects on the national health systems and the global economy. This situation has led to searches to control the devastating effects of the pandemic and to provide social immunity against the COVID-19 disease. Today, many vaccines have been developed against COVID-19 using different health technologies.[1,2]

COVID-19 vaccines have been very beneficial in preventing infectious diseases, reducing mortality, and controlling the pandemic.[3] In addition, the route of the COVID-19 pandemic and how effective the vaccines will show against possible new variants are still a matter of curiosity. [4]

The main goal of COVID-19 vaccine applications is to provide a strong immune response as a result of stimulating the immune system. The immune response that occurs after vaccination includes components such as innate immunity, cellular immunity, humoral immunity, and cytokine response.[5,6] Serological detection of the antibody response resulting from humoral immunity is a convenient method that can be used to determine the presence and level of the immune response after vaccination.[7] In serological tests, using purified SARS-CoV-2 proteins such as the nucleocapsid protein (NCP) and spike (S) protein, or more specific subregions such as the S1 and S2 subunits or the receptor binding domain (RBD) in the S1 subunit, the presence of virus-specific IgM, IgG or IgA antibodies in the blood can be determined.[8]

Many test kits have been developed to detect antibodies against SARS-CoV-2. The most important disadvantage of the antibody tests is the inconsistency between the results of the kits in which antibodies against different viral proteins were investigated.[9] In general, it is stated that a better antibody response occurs thanks to COVID-19 vaccines, especially after the second dose. In addition, it is clarified that the immune response that occurs after vaccination is stronger than the immune response that occurs with a natural infection and that people who have had COVID-19 will also benefit from vaccination.[9] On the other hand, factors such as individual characteristics such as age, gender, presence of comorbidities, genetics and nutrition, vaccine type, presence of adjuvant, and vaccine dose may affect the effectiveness of the immune response and the duration of vaccine-induced immunity.[10,11]

Since healthcare workers have a high risk of encountering COVID-19 infection, these individuals have been given priority in the CoronaVac (Sinovac, China) vaccination program in our country. In this study, it was aimed to investigate the relationship between the antibody response after the second dose of vaccination, and gender, age, body mass index, smoking, the presence of COVID-19 history before vaccination, presence of diabetes mellitus, and hypertension in healthcare workers. In addition, it is planned to observationally examine the third and fourth-dose vaccination approaches of the participants and the protection of the antibody levels detected after two doses of vaccination against COVID-19.

Material and Methods

One hundred and fifty-six healthcare professionals were included in this prospective study between the study period, January- December 2021. Two doses of the CoronaVac vaccine were administered to healthcare workers in January and February 2021. The third and/or fourth dose vaccination and COVID-19 status of healthcare workers whose NCP and S protein antibody levels were detected on the 30th day after the second dose of vaccination were followed up to two years until 2023.

3-5 mL of the blood sample taken from volunteer health workers participating in the research were separated into serum by centrifugation. The presence of NCP and S protein antibodies in serum samples were analyzed with Elecsys Anti-SARS-CoV-2 (Roche Diagnostics, Switzerland) and Elecsys Anti-SARS-CoV-2 S kit (Roche Diagnostics, Switzerland) test kits, respectively using the Electro-chemiluminescence Immune Assay (ECLIA) method in the Cobas e601 (Roche Diagnostics, Switzerland) device following the manufacturer's recommendations.

The presence of NCP antibody in serum samples was qualitatively evaluated using the Elecsys Anti-SARS-CoV-2 kit (Roche Diagnostics, Switzerland). IgM and IgG-type antibodies developed against NCP were measured according to the Cut-Off Index (COI) value in accordance with the manufacturer's recommendations. COI < 1; is rated "*nonreactive*", and COI ≥1; is rated as "*reactive*". All antibodies including IgG, against the RBD region of the S protein, were evaluated quantitatively using the Elecsys Anti-SARS-CoV-2 S kit (Roche Diagnostics, Switzerland). In this assessment, results determined as greater than 0.8 U/mL were considered "*reactive*", and the highest antibody value was measured as 250 U/mL by the device. The values measured above 250 U/mL were accepted as >250 U/mL.

Statistical analysis was performed using SPSS 22.0 (IBM Co., USA). In the assessment of the results, antibody levels were grouped as 1-125, 126-250, and >250 U/mL and evaluated as the percent. The difference between the groups was evaluated with the $\chi2$, Fisher's exact, Mann-Whitney U, and Kruskal-Wallis tests in accordance with the convenience. The value of *p <0.05* was considered statistically significant.

Ethical Approval

The study protocol was approved by the Sivas Cumhuriyet University Clinical Research Ethics Committee (Date: 21.12.2020, Decision number: 2020-12/03). Each stage of the research was performed per the ethical standards specified in the 1964 Declaration of Helsinki and its later amendments.

Results

A total of 156 health workers, 77 (49.4%) women, and 79 (50.6%) men, were included in this study. The ages of the participants are between 21-59 years. The mean age of the participants was 36.76±8.9. The mean age of female participants was 36.55±8.3, and of male participants was 36.96±9.5. Among all the participants sixty (38.5%) of them had experienced COVID-19 infection before they were given two doses of the CoronaVac vaccine and 96 (61.5%) had no history of the previous COVID-19 infection. Thirty-three (55%) of the healthcare workers who had COVID-19 infection were female, and 27 (45%) were male. Among participants who had COVID-19 infection before two doses of CoronaVac vaccination, measurable NCP-antibody levels were detected (>1) in 52 cases (86.7%), while measurable NCP antibody levels were not detected in eight cases (13.3%). On day 30, after two doses of CoronaVac vaccination, a total of 155 (99.4%) participants had measurable levels of S antibody (>0.8), and a total of 135 participants (86.5%) had measurable levels of NCP antibodies (>1).

In this study, S antibody was found to be negative after two doses of the CoronaVac vaccine only in one healthcare worker. This person was a 41-year-old female health worker with a body mass index of 29.2. She does not smoke or has no comorbidities. She was not infected with COVID-19 before the CoronaVac vaccination. In this healthcare worker COVID-19 PCR test positivity was detected after the fourth dose of vaccination, whose third and fourth doses were vaccinated with the Pfizer-BioNTech vaccine (BNT162b2).

Table 1 shows the distribution of S antibody levels on the 30th day after two doses of CoronaVac vaccination of healthcare workers included in this study, according to demographic characteristics such as gender, age groups, body mass index, smoking, and comorbidity (Table 1).

Table 1. Distribution of S antibody levels according to demographic characteristics at day 30 after vaccination of the healthcare worker with two doses of CoronaVac.

		S antibody level (n=155)			
		1–125 U/mL	126–250 U/mL	>250 U/mL	
	n	n (%)			p
COVID-19 infection status before vaccination					
Yes	60	8 (13.3)	8 (13.3)	44 (73.4)	0.000
No	95	38 (40)	20 (21.1)	37 (38.9)	
Gender					
Woman	76	13 (17.1)	13 (17.1)	50 (65.8)	0.001
Man	79	33 (41.8)	15 (19)	31 (39.2)	
Age groups					
20-29	50	9 (18.0)	7 (14.0)	34 (68.0)	
30-39	43	12 (27.9)	8 (18.6)	23 (53.5)	0.123
40-49	42	17 (40.5)	9 (21.4)	16 (38.1)	
50-59	20	8 (40.0)	4 (20.0)	8 (40.0)	
Body mass index					
Normal (18.5-24.9)	92	30 (32.6)	14 (15.2)	48 (52.2)	
Overweight (25-29.9)	52	13 (25.0)	12 (23.1)	27 (51.9)	0.773
Obesity (≥30)	11	3 (27.3)	2 (18.2)	6 (54.5)	
Smoking					
Yes	83	32 (38.5)	17 (20.5)	34 (41.0)	0.008
No	72	14 (19.4)	11 (15.3)	47 (65.3)	
Diabetes mellitus					
Yes	16	5 (31.3)	3(18.7)	8 (50.0)	0.982
No	139	41 (29.5)	25 (18)	73 (52.5)	
Hypertension					
Yes	18	9 (50.0)	4 (22.2)	5 (27.8)	0.068
No	137	37 (27.0)	24 (17.5)	76 (55.5)	
NCP antibody level					
Nonreactive	20	16 (80.0)	3 (15.0)	1 (5.0)	0.000
Reactive	135	30 (22.2)	25 (18.5)	80 (59.3)	
Total	155	46 (29.7)	28 (18)	81 (52.3)	

Table 2. Vaccination status of healthcare workers

Vaccine	First dose	Second dose	Third dose	Fourth dose
		n (%)		
CoronaVac	156 (100.0)	156 (100.0)	2 (1.3)	0 (0.0)
Pfizer-BioNTech	0 (0.0)	0 (0.0)	150 (96.1)	110 (70.5)
Unvaccinated person	0 (0.0)	0 (0.0)	4 (2.6)	46 (29.5)

S and NCP antibody levels in blood samples taken 30 days after the second dose of vaccination were compared in this study. NCP antibody was found nonreactive; in 16 (34.8%) of the 46 healthcare workers with S antibody levels between 1–125 U/mL; in 3 (10.7%) of 28 healthcare workers with S antibody levels between 126–250 U/mL, and one (1.2%) of 81 healthcare workers with S antibody levels >250 U/mL.

The first, second, third, and fourth dose vaccination statues of the healthcare workers included in this study are given in Table 2. 152 (97.4%) of the participants had the third dose and 110 (70.5%) of the participants had the fourth dose of the vaccine. 46 healthcare workers (29.5%) did not vaccinate with the fourth dose (Table 2).

The distribution of antibody levels and demographic characteristics of healthcare workers with COVID-19 infection after two doses of CoronaVac vaccination according to their history of COVID-19 infection before vaccination is presented in Table 3.

A total of 48 (30.8%) healthcare workers included in the study had COVID-19 infection after two doses of vaccination. In 14 (29.2) of these people, COVID-19 infection history was detected before two doses of vaccination, and COVID-19 infection was diagnosed by PCR test. It was determined that 34 (70.8) healthcare workers did not have COVID-19 infection before vaccination (Table 3).

In this study, it was specifically found that people who had COVID-19 infection before vaccination had higher NCP antibodies after two doses of vaccination (*p=0.013*). On the other hand, there was no difference in S antibody levels after two doses of vaccination between groups with and without COVID-19 infection (*p=0.183*) (Table 3).

Table 3. Distribution of antibody levels and demographic characteristics of healthcare workers who had COVID-19 infection after two doses of vaccination, according to their history of having COVID-19 infection before vaccination.

		Infected COVID-19	Not Infected COVID-19	p
	n	n (%)		
COVID-19 period				
After the second dose	9	4 (28.6)	5 (14.7)	
After the third dose	21	5 (35.7)	16 (47.1)	0.515
After the fourth dose	18	5 (35.7)	13 (38.2)	
***Spike antibody**				
<1 U/mL	1	0 (0)	1 (2.9)	
1–125 U/mL	13	2 (14.3)	11 (32.4)	0.183
125-250 U/mL	8	1 (7.1)	7 (20.6)	
>250 U/mL	26	11 (78.6)	15 (44.1)	
***Nucleocapsid antibody**				
<1 U/mL	10	1 (7.1)	9 (26.5)	
1–125 U/mL	30	7 (50)	23 (67.6)	0.013
125-250 U/mL	7	5 (35.7)	2 (5.9)	
>250 U/mL	1	1 (7.1)	0 (0)	
Gender				
Woman	25	6 (42.9)	19 (55.9)	0.412
Man	23	8 (57.1)	15 (44.1)	
Age groups				
20-29	15	5 (35.7)	10 (29.4)	
30-39	14	5 (35.7)	9 (26.5)	0.407
40-49	13	4 (28.6)	9 (26.5)	
50-59	6	0 (0)	6 (17.6)	
Total	48	14 (29.2)	34 (70.8)	

Discussion

Investigating the antibody response to SARS-CoV-2 can provide important data to understand whether people have been infected with this virus before, to diagnose a possible infection, and to determine the impact of the vaccine in case of vaccination.[9] In this study, the relationship between the antibody response after the second dose of vaccination in healthcare workers and history of COVID-19 before vaccination, gender, age, body mass index, smoking, and presence of chronic disease was investigated.

In the literature, it is reported that the immune response that occurs after vaccination is higher than the immune response that occurs with natural infection and that people who have had COVID-19 infection will also benefit from the vaccine.[9] In our study, it was found that the people who had COVID-19 infection before two doses of vaccination had higher antibody levels compared to people without a history of COVID-19 (*p=0.000*). This result is compatible with the literature.

A study conducted by Uysal et al.[12] showed that among 314 healthcare workers S antibody levels were above >250 U/mL in 56% of women and 44% of men after two doses of CoronaVac vaccination. However, they reported

that no significant difference was found between the genders (*p=0.111*). In another study, no difference was found between genders in terms of antibody levels after vaccination.[13] However, in our study, higher S antibody levels were detected in women compared to men after two doses of vaccination (*p=0.001*). Consistent with our study, in a study conducted by Akaret al.[11] in Türkiye, it was reported that the antibody response was found to be significantly higher in women after vaccination. This difference between the gender is considered as usual since the difference in the duration of infection in women and men may affect the humoral immune response after vaccination.[14]

Germinal centers in lymph nodes are highly functional structures for antibody response formation after vaccination. As a result of the decrease in the size and functions of the germinal center with aging, it causes weakening of the humoral response and a decrease in antibody levels after vaccination.[15] In the study of Uysal et al,[12] it was reported that higher antibody levels were detected in the 30-39 age group (38.5%) compared to other age groups. In another study by Seyahi et al,[16] lower antibody levels were observed after two doses of vaccination in people over the age of 65 compared to hospital workers of younger ages. In our study, no statistically significant relationship was found between age groups and antibody levels (*p=0.123*). However, the detection of higher antibody levels in the ages of 20-29 and 30-39 compared to other age groups in our study is similar with the literature.

It has been reported that smoking and high body mass index negatively affect the humoral response to COVID-19 vaccines and cause low antibody response.[11,17,18] In our study, in concordance with the literature, it was determined that smoking had a negative effect on the antibody response after vaccination (*p=0.008*). After two doses of vaccination, antibody levels were >250 U/mL in 65.3% (47/72) of non-smokers and 41% (34/83) of smokers.

Soegiarto et al.[19] pointed out to the associated hypertension with low antibody levels after CoronaVac vaccination. Consistent with this study, lower S antibody levels were found in healthcare workers with a history of hypertension in our study (*p=0.068*). While >250 U/mL of S antibody levels were observed in 27.8% of patients with a history of hypertension, S antibody levels >250 U/mL were detected in more than half (55.5%) of those who did not have this history.

The limitation of our study was that the Elecsys Anti-SARS-CoV-2 S-kit could not be obtained before two doses of CoronaVac vaccination of healthcare workers, so the levels of S antibody before vaccination could not be determined. In addition, only the humoral antibody response of the vaccine was revealed in our study; thus, no information could be given about cellular immunity. In addition, the participants preferred different vaccines for the third and fourth doses. In our study, we could not investigate how and to what extent different vaccine preferences affect the effectiveness of antibody response

therefore this could be considered a limiting situation as well.

Conclusion

As a result, the CoronaVac®, which is an inactivated SARS-CoV-2 vaccine, provides an adequate S antibody response as 99.4%, 30 days after two doses of vaccination in healthcare workers aged 21-59 years. The antibody response after two doses of inactivated CoronaVac® vaccination is higher in women, young people, non-smokers and people previously infected with SARS-CoV-2. The exact protective effect of the antibody levels obtained with the vaccine are in practice will be better understood in the periods when the number of new cases increases.

Declaration of Conflict of Interest

The authors declared no potential conflicts of interest concerning this article's research, authorship, and publication.

Funding

The authors received no financial support for this article's research, authorship, and publication.

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