



## Sleep Quality Affects Humoral Response in Recipients of Two-Dose Sinovac Vaccines

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### Abstract

To curb the COVID-19 pandemic, the government distributed Sinovac vaccines. Sleep mediates immune function, including post-vaccination antibody response. This study aimed to analyze whether there was a difference in post-vaccination antibody levels in Sinovac vaccine recipients with poor and good sleep quality. This study used analytical observations of recipients of the two-dose Sinovac vaccine in 2021. Primary data included age, sex, the Pittsburgh Sleep Quality Index (PSQI) questionnaire, and post-vaccination IgG-SARS-CoV-2 antibody levels. The PSQI and IgG SARS-CoV-2 antibody levels were measured a month after the second vaccination. Participants with non-reactive antibody levels before the first vaccination were included, and participants with incomplete data were excluded. The Mann-Whitney test was used to find associations between sleep quality and post-vaccination IgG SARS-CoV-2 levels. The univariate analysis showed that of 54 participants, 37 (68.5%) were male, and 28 (51.9%) had poor sleep quality. 15 participants (27.78%) were in the 36-45 age group, and median antibody levels in participants who received the second Sinovac Vaccine was 223.5 (199.01) units/mL. Post-vaccination IgG SARS-CoV-2 antibody levels were significantly associated with sleep quality ( $p=0.036$ ).

### Introduction

The effect of sleep on the immune system is essential to characterize in light of the COVID-19 (coronavirus disease 2019) pandemic (Cucinotta and Vanelli, 2020), which has caused changes in daily activities and sleep dysregulation. The effect of sleep on the immune system is significant to characterize in light of the COVID-19 pandemic (Cucinotta and Vanelli, 2020). It can cause changes in daily activities and sleep dysregulation. A meta-analysis found that the global prevalence of sleep disturbances during the COVID-19 pandemic was 40.49% and increased during the lockdown (Jahrami et al., 2022). Several studies show that sleep mediates immune function, including post-vaccination antibody levels. At night, during sleep, circulating B cells and T cells move into the lymph nodes, where the immune system recognizes antigens such as viruses, and an

immune response is then formulated (Comas et al., 2017; Schmitz, van der Werf and Lammers-van der Holst, 2022). Short sleep duration has been found to affect the human body's immune system (Schmitz, van der Werf, and Lammers-van der Holst, 2022). Individuals with poor sleep quality are also more susceptible to infectious diseases (Kow and Hasan, 2021). To elucidate, a study found that individuals who slept  $\leq 5$  hours a night were more susceptible to pneumonia (Patel et al., 2012). Another study found that individuals who slept  $< 7$  hours a night were more susceptible to colds (Cohen et al., 2009). Other researchers have also found an association between short sleep duration and lower antibody levels post-hepatitis B vaccination (Prather et al., 2012).

Sleep affects the immune system through the hypothalamic-pituitary-adrenal axis (HPA axis). The paraventricular nucleus (PVN) in the

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hypothalamus releases corticotropin-releasing Hormone (CRH), which induces the release of adrenocorticotrophic hormone (ACTH), thus stimulating the adrenal glands to release cortisol, a stress hormone. Physiologically, sleep suppresses the HPA axis, which subdues cortisol release. Therefore, sleep disturbances increase cortisol levels (Balbo, Leproult, and Van Cauter, 2010), reduce CD4+ and Natural Killer cells (Asif, Iqbal, and Nazir, 2017).

COVID-19 caused by acute respiratory syndrome coronavirus 2 (SARS-CoV-2) (Cucinotta and Vanelli, 2020) is highly infectious and has several risk factors, including advanced age and co-morbidities such as type I and II diabetes and hypertension (Wolff et al., 2021). Globally, as of August 2021, at least 208,470,375 individuals were infected, and COVID-19 is associated with 4,377,979 deaths. In Indonesia, at least 3,892,479 individuals were infected, and symptomatic patients present most often with cough and fever (Setiadi et al., 2022). To curb the COVID-19 pandemic, the Indonesian government provided Sinovac vaccines in December 2020, a whole inactivated virus that reduces transmission rates and protects against disease severity. A study in Brazil found that the Sinovac Vaccine at 14 to 30 days after administration of the second dose had an efficacy of 55% against symptomatic COVID-19 disease and 82.1% against severe infection (Cerqueira-Silva et al., 2022). So far, the effect of gender on post-vaccination antibody levels in recipients of the Sinovac Vaccine is conflicting, but age seems to mediate humoral response (Heriyanto et al., 2021; Farid, Herrera-Urbe and Stevenson, 2022). The main aim of this study is to determine whether sleep quality affects post-vaccination IgG SARS-CoV-2 antibody levels in recipients of the Sinovac vaccine. To our knowledge, this study is the first to investigate differences between sleep quality and post-vaccination SARS-CoV-2 IgG antibody levels in Indonesia.

## Method

This study uses an analytical observational approach towards data (age, gender, sleep quality, and post-vaccination IgG SARS-CoV-2 levels) from a cross-sectional conducted in 2021 at a vaccine center in Jakarta. Participants came

from the general population and received two doses of the Sinovac vaccine. Antibody levels were measured before the first vaccination and a month after the second vaccination. Participants with non-reactive antibody levels before the first vaccination were included, and participants with incomplete data were excluded. IgG (Immunoglobulin G) antibodies against the receptor binding protein on the Spike (S) protein of SARS-CoV-2 determination is by the electro-chemiluminescence immunoassay (ECLIA) method through the Elecys® Anti-SARS-CoV-2 S assay. It has a sensitivity of 98.8% and a specificity of 99.98%. Results were expressed in units/mL, with a range of 0.4-250 units/mL, and were considered reactive if  $\geq 0.8$  units/mL (Taffertshofer et al., 2022).

We gave a general questionnaire asking participants' characteristics, such as age, gender, and the Pittsburgh Sleep Quality Index. The PSQI is widely used to evaluate sleep quality over the past month and comprises seven components (subjective sleep quality, sleep latency, duration, habitual sleep efficiency, sleep disturbances, use of sleeping medications, and daytime dysfunction). The scores from each component are added and given a global score with a range of 0-21, with a cut-off score of 5. Good sleep quality is defined by a global PSQI score of  $\leq 5$ , while poor sleep quality by a global PSQI score of  $> 5$ . The PSQI has a Cronbach alpha score of 0.79, a validity of 0.89, a sensitivity of one, and a specificity of 0.81 (Ikbal Zendi Alim, 2015).

Data analysis is by the Statistical Package for the Social Sciences (SPSS), version 25 (SPSS Inc., Chicago, IL, USA). Data were considered normal if the Kolmogorov-Smirnov criteria were fulfilled. Continuous variables were expressed as mean ( $\pm$  SD) and also as median (interquartile [IQR]) range if skewed. Sleep quality was dichotomized as good (PSQI  $\leq 5$ ) and poor (PSQI  $> 5$ ). Data analysis is by the Mann-Whitney and Kruskal-Wallis tests. Values of  $p < 0.05$  were considered statistically significant.

## Result and Discussion

Initial data from 85 participants were available. However, only 64 participants fulfilled the inclusion criteria, serologically

non-reactive antibody titers before the first vaccination, and a further 10 participants were excluded due to incomplete data. Hence, data from 54 participants were available to analyze.

**TABLE 1.** Characteristics of Study Participants According to Gender, Age, Sleep Quality, and Post-Vaccination IgG SARS-CoV-2 levels in Recipients of the Second Dose of the Sinovac Vaccine

	Total (n)	Percentage (%)
Gender		
Male	37	68.5*
Female	17	31.5
Age (Mean $\pm$ SD)		39.74 $\pm$ 11.98 years
17-25	10	18.52
26-35	10	18.52
36-45	15	27.78*
46-55	14	25.93
56-65	5	9.23
Sleep quality		
Good	26	48.1
Poor	28	51.9*
Post vaccination IgG SARS-CoV-2 levels (Mean $\pm$ SD) [Median (Interquartile range)]; Min-Max units/mL		163.72 $\pm$ 97.53 [223.5 (199.01)]; 7.2-250

\*Highest percentage

Source: Primary Data, 2021

Among 54 study participants, 68.5% were male, 27.78% belonged to the age group 36-45 years with mean age of 39.74 ( $\pm$  11.98) years, 51.9% had poor sleep quality, and mean post-vaccination IgG SARS-CoV-2 levels were 163.72  $\pm$  97.53 units/mL [Median (Interquartile range) = 223.5 (199.01)] (Table 1). Most participants included in this study were male, and all were of productive age. The percentage of participants with poor sleep quality (PSQI > 5) as per a study which found that 59.5% of Indonesians had poor sleep quality during the COVID-19 pandemic (Argo et al., 2021). A meta-analysis concluded that the prevalence of sleep disturbances during the pandemic was influenced by the rate of disease transmission and government policies such as enforcing lockdowns, which caused changes in bedtime and wake time. Restriction of outdoor activities and social isolation also increased social media and technology use, and the blue light emitted

from gadgets has been found to disturb the circadian rhythm (Jahrami et al., 2022).

The Sinovac vaccine was successful in producing reactive antibody levels ( $\geq$  0.8 units/mL). A month after the second vaccination, IgG SARS-CoV-2 levels increased to 223.5 (199.01) units/mL. The lowest measured post-vaccination IgG SARS-CoV-2 level was 7.2 units/mL, and the highest was 250 units/mL (Table 1). The results of this study are similar to one whose participants received the BNT162b2 (Pfizer/BioNTech) vaccine, and as much as 90% of participants had reactive antibody levels after 21 days after vaccination (Ward et al., 2022). The Sinovac vaccine applies the traditional whole inactivated virus method, whereas the BNT162b2 (Pfizer/BioNTech) vaccine uses a novel mRNA method capable of producing higher levels of antibodies. However, the Sinovac Vaccine is easier to transport and store (Lim et al., 2021).

**TABLE 2.** Association Between Gender and Post-Vaccination IgG SARS-CoV-2 Levels

Post vaccination IgG SARS-CoV-2 levels	N	Mean	Std. Deviation	Median (IQR)	<i>p</i>
Gender					
Male	37	162.131622	100.852346	224.3 (202.635)	0.661
Female	17	167.177647	92.7747521	209.1 (171.62)	

Source: Primary Data, 2021

The Mann-Whitney test found no significant association between gender and post-vaccination IgG SARS-CoV-2 levels ( $p=0.661$ ). However, female participants had a mean antibody level of  $167 \pm 92,77$  units/mL and males  $167 \pm 92,77$  units/mL (Table 2). The results of this study are per a study on Greek healthcare workers, which found no association between gender and antibody levels in 268 participants (79.9% female), when measured 30 days after the second dose of BNT162b2 (Pfizer/BioNTech) vaccines [(Median (Interquartile range) = 1288.00 (1376.95 units/mL)] (Michos et al., 2021), and in a study investigating whether gender affects antibody levels in recipients of the Sinovac vaccine in Bahrain (Farid, Herrera-Urbe, and Stevenson, 2022). Another study found that of 439 participants (65.8% female), mean antibody levels measured 3-4 weeks after the second dose of BNT162b2 (Pfizer/BioNTech) were higher in females than males. However, this association was significant only in the 51-60 age group (Anastassopoulou et al., 2022).

A cross-sectional study also found that post-vaccination antibody levels after administration of BNT162b2 (Pfizer/BioNTech) vaccine were higher in females than males and was statistically significant (Ward et al., 2022). A prospective study by Tsverava et al. (2021) found that gender differences in antibody levels after infection with SARS-CoV-2 depended on the fragments of the Spike protein measured, such as the S1 protein, S2 protein, and the

receptor binding domain (RBD). The RBD is found on the S1 fragment and is crucial in facilitating viral entry into target cells. Tsverava et al. (2021), found a statistically significant relationship between gender and antibody levels against the S1 protein. However, the relationship was only significant on a one-tailed test against the RBD, and no relationship was found when measured against the S2 protein. Therefore, differences in results regarding gender's influence on antibody levels may be due to the different antigens contained in the Sinovac and BNT162b2 (Pfizer/BioNTech) vaccines. As mentioned above, the Sinovac Vaccine uses the whole inactivated virus method. The BNT162b2 (Pfizer/BioNTech) vaccine uses the mRNA method, which causes the human body to produce only a fragment of the Spike protein. Little research has been done on how gender influences antibody response (Anastassopoulou et al., 2022). However, estrogen increases B cell production, and post-vaccination antibody levels negatively correlate with serum testosterone (Athanasίου et al., 2022). Females also have two X chromosomes associated with higher counts of lymphocytes (Tsverava et al., 2021). The Kruskal-Wallis test found no statistically significant association between age and post-vaccination IgG SARS-CoV-2 antibody levels ( $p=0.154$ ). Age was categorized into five groups according to the 2009 classification provided by the Indonesian Ministry of Health.

**TABLE 3.** Association Between Age and Post-Vaccination IgG SARS-CoV-2 Levels

	Age		Post vaccination IgG SARS-CoV-2 levels					
	Range	N	Mean	Min	Max	Std. Deviation	Median (IQR)	<i>p</i>
Late adolescence	17-25	10	224.079000	31.69	250	68.8097891	250 (10.225)	
Early adulthood	26-35	10	121.356	18.63	250	98.2999112	99.130 (203.1175)	
Late adulthood	36-45	15	152.280	14.44	250	95.0139	136.3 (193.53)	0,154
Early elderhood	46-55	15	155.17	7.2	250	112.43	236.35 (233.03)	
Late elderhood	56-65	5	185.992	97.16	250	77.52	228.7 (147.37)	

Source: Primary Data, 2021

The lowest post-vaccination IgG SARS-CoV-2 levels [99.13 (203.1175) units/mL] were in the second age group (26-35 years). The lowest antibody response mounted (7.2 units/mL) was found in the fourth age group (46-55 years). However, the highest antibody level [Mean=  $224 \pm 68.8$  units/mL, Median

(Interquartile range)= 250 (10.225) units/mL] was found in the youngest age group (17-25 years) (Table 3). Michos et al. (2021) also found that IgG SARS-CoV-2 levels were higher in individuals < 60 years than in individuals  $\geq$  60. Anastassopoulou et al. (2022) had similar results. They found that after administration of

the BNT162b2 (Pfizer/BioNTech) vaccine, the highest antibody levels were in the youngest age group (21-30 years), and the lowest antibody levels were found in the 31-40 age group, which is similar to the results of this study (26-35 age group). Ward et al. (2022), found that antibody levels in the 70-79 age group were significantly

lower when compared to the 18-29 age group. Although not much research has been done on how age affects humoral response, increasing age reduces CD4+ response and is associated with a shift towards the anti-inflammatory interleukin-2 and interleukin-10 (Zimmermann and Curtis, 2019).

**TABLE 4.** Association Between Sleep Quality and Post-Vaccination IgG SARS-CoV-2 Levels

Post vaccination IgG SARS-CoV-2 levels		N	Mean	Std. Deviation	Median (IQR)	<i>p</i>
Sleep quality	Good	26	194.924231	81.3427312	250 (118.4)	0.036
	Poor	28	134.745000	103.625820	118.65 (227.36)	

Source: Primary Data, 2021

The Mann-Whitney test results showed a statistically significant association between sleep quality and post-vaccination IgG SARS-CoV-2 levels ( $p = 0.036$ ). Antibody levels were lower [Mean =  $134.74 \pm 103.63$  unit/mL, Median (Interquartile range) = 118.65 (227.37)] in the group with poor sleep quality (PSQI > 5) than in the group with good sleep quality [Mean =  $194.92 \pm 81.34$  unit/mL, Median (Interquartile range) = 250 (118.4)]. (Table 4). In recipients of the influenza vaccine, participants with poor sleep quality measured by the PSQI had lower antibody responses after administration of the vaccine ( $p < 0.001$ ) (Taylor et al., 2017). In Greece, study results found that participants with PSQI > 5 had lower IgG SARS-CoV-2 levels in recipients of the BNT162b2 (Pfizer/BioNTech) vaccine ( $p < 0.05$ ) (Athanasίου et al., 2022). Several studies have found that other parameters of sleep, such as sleep duration, are associated with post-vaccination antibody levels. Sleep diaries and actigraphy are particularly useful in measuring sleep duration and individual variations in sleep duration before and after vaccination (Prather et al., 2012; Prather et al., 2010). According to the results of a study on recipients of the hepatitis B vaccine, acute sleep deprivation before administration was associated with lower antibody levels (Lange et al., 2003). Two nights before influenza vaccines were administered, electronic sleep diaries were used to measure sleep duration, and researchers found that shorter sleep duration was associated with lower antibody levels (Prather et al., 2021). In contrast, Athanasίου et al. (2022), measured sleep duration two nights

before and a night after administration of the BNT162b2 (Pfizer/BioNTech) vaccine but found no relationship between sleep duration and post-vaccination antibody levels. During sleep, T cells introduce antigens to the body's immune system, and B cells produce antibodies (Schmitz, van der Werf, and Lammers-van der Holst, 2022). Sleep also reduces cortisol levels, which is anti-inflammatory and reduces the immune system's responsiveness (Lange et al., 2011). Sleep increases interleukin-12, a cytokine that supports T-cell and B-cell differentiation (Lange et al., 2003).

## Conclusion

All participants were able to produce reactive IgG SARS-CoV-2 levels after receiving two doses of the Sinovac Vaccine, and the highest average levels were in the 17-25 age group. Sleep quality was associated with post-vaccination IgG SARS-CoV-2 levels in recipients of the Sinovac Vaccine ( $p = 0.036$ ). Educational institutes focusing on research and community outreach should screen antibody levels periodically and investigate their associations with physical and mental well-being. The general population should also be enlightened on the benefits of receiving COVID-19 vaccines.

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