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RESEARCH ARTICLE

EVALUATING THE DURABILITY OF HUMORAL IMMUNITY AGAINST SARS-COV-2 SPIKE PROTEIN 18 MONTHS AFTER VACCINATION: A RANDOMIZED SEROLOGICAL STUDY

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ABSTRACT

One of the most important ways to lessen the effects of the COVID-19 pandemic, which has caused an unprecedented global health crisis, is through vaccination. Despite the controversy surrounding COVID-19 vaccines, Covishield has been India's most popular vaccine. One and a half years after vaccination, this study examines the persistence of antibodies against COVID-19, particularly those produced by Covishield. 350 participants in total, equally split between the sexes, were chosen for this analysis. Serious public health consequences may result from the presence of antibodies years after vaccination. Our results demonstrate the effectiveness of vaccination, as 68% of samples had adequate antibody titers (>0.80 U/mL) against COVID-19. The concurrent emergence of neurological, respiratory, and cardiovascular adverse effects in those who mounted a sufficient antibody response was a noteworthy finding, though. On the other hand, individuals who were unable to produce adequate antibody levels frequently had a significant medical history, indicating that a serious illness could impair the immune response to vaccination. The significance of additional research into the variables influencing antibody development and vaccine effectiveness is underscored by these findings, which show the delicate relationship between vaccine efficacy and individual health status.

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INTRODUCTION

The highly contagious, positive-sense, single-stranded RNA virus known as SARS-CoV-2 (Severe Acute Respiratory Syndrome Coronavirus 2) has spread quickly worldwide. When this virus infects a person, the result is COVID-19 (coronavirus disease 2019), which can cause a variety of symptoms such as fever, coughing, chest pain, and, in extreme situations, respiratory distress syndrome. [1, 2]. Eleven new COVID-19 vaccines have been developed and proven effective; several more are undergoing Phase III clinical trials. However, more vaccines are still required due to the exceptional demand for vaccines worldwide and the increasing impracticability of conducting placebo-controlled efficacy trials. [3]. To mitigate the COVID-19 pandemic, effective and safe vaccines are essential. To accelerate the development of current SARS-CoV-2 vaccines and prepare the way for their possible widespread deployment, the literature currently in publication describes developments in the development of SARS-CoV and Middle East respiratory syndrome coronavirus (MERS-CoV) vaccines. Inactivated vaccines, live attenuated vaccines, vector-based vaccines, RNA-based vaccines, DNA-based vaccines, protein subunit vaccines, and virus-like particle (VLP) vaccines are the distinct categories into which the World Health Organization (WHO) divides COVID-19 vaccines that are being examined or approved for clinical trials[4]. The spike proteins facilitate viral entry into host cells. The S1 and S2 subunits make up these spike proteins. The S1 subunit facilitates viral attachment and allows the virus to identify host receptors. The S2 subunit of the spike proteins aids in the fusion of the virus with the membrane of the host cell. The M proteins are the most abundant proteinaceous component of the virion and give the viral envelope its final shape. They are essential to the coordination of viral assembly[5]. Transmembrane domains, a shortened amino-terminal segment, and an extended carboxy-terminal segment make up the M protein. The tiny membrane polypeptide known as the envelope protein serves as an ion channel. The E protein is made up of a carboxy-terminal domain, a large hydrophobic transmembrane domain, and a brief hydrophilic amino-terminal segment. The N protein is essential for controlling transcription, viral RNA synthesis, and the control of metabolic activities in infected cells. These proteins encapsulate the genomic RNA to protect it. SARS-CoV-2 takes an average of 5.1 days to incubate. Within 11.5 days, symptoms start to appear. Between 17.9% and 33.3% of patients are thought to be asymptomatic. Pyrexia, cough, dyspnea, and, less commonly, sore throat, dysgeusia, anorexia, nausea, respiratory distress, cephalgia, malaise, myalgias, and diarrhea are the most common clinical manifestations of infection[6]. To prevent disease, vaccines are made up of toxins, attenuated or inactivated microorganisms, or other biological preparations that contain antibodies, lymphocytes, or mRNA. Vaccines provide both active acquired and passive immunity against a particular harmful agent by inducing the immune system to target that agent. Although some vaccines are given orally or through the nose, injections are the most common method of administration[7]. In December 2020, the first COVID-19 mass vaccination campaign got underway. Using data from scientific trials and production procedures, vaccines are subjected to a thorough evaluation to

guarantee compliance with relevant safety and effectiveness standards. The assessment weighs the possible advantages of using the product against any associated risks, as well as the threat posed by the emergency[8].There are three vaccines available in India: Sputnik V, Covaxin, and Covishield. On January 16, 2021, the Serum Institute of India approved the use of Covaxin and Covishield in an emergency. The SARS-CoV-2 S protein is encoded by the recombinant, non-replicating chimpanzee adenovirus vector Covishield, which is generated in genetically altered human embryonic kidney (HEK) 293 cells[9]. Through the development of memory T-cells and B-cells, this vaccine stimulates the production of antibodies and produces an immune response. In addition to L-histidine, L-histidine hydrochloride monohydrate, magnesium chloride hexahydrate, polysorbate 80, ethanol, sucrose, sodium chloride, disodium edetate dihydrate (EDTA), and water for injection, Covishield contains a recombinant replication-deficient chimpanzee adenovirus vector that encodes the SARS-CoV-2 spike (S) glycoprotein[10]. The development of immunity against SARS-CoV-2 infection is significantly influenced by antibody responses following COVID-19 vaccination. The body frequently produces protective antibodies when the COVID-19 virus is introduced into the human system, either naturally or through vaccination. Immunoglobulin IgM antibodies typically form in 7–9 days, while IgG antibodies appear about 2 weeks after viral exposure[11]. The following techniques can be used to detect antibodies in blood samples: ex vivo interferon gamma ELISpot assays, hemagglutination tests, surrogate virus neutralization tests, and enzyme-linked immunosorbent assays[12].

METHODOLOGY

Using predetermined inclusion and exclusion criteria, this study was carried out in a community setting. 350 samples in all, including both male and female samples, were enrolled. The second dose of the vaccine had to be administered 1.5 years before sample collection to meet the inclusion criteria. Male and female samples were equally distributed, and participants were chosen from the 18–55 age range. Participants with a history of COVID-19 symptoms were not allowed to participate in the study. Each participant signed a consent form indicating their informed consent. The JJTU Human Ethics Committee gave its approval to the study. Using a specific data collection form, information about the samples was gathered. Based on the inclusion and exclusion criteria, samples were chosen at random, sent to the laboratory for sample collection, and required to sign an informed consent form.To find out whether total IgG antibodies were present, the specimen was examined at Laboratory using an electrochemiluminescence immunoassay. This specific assay, which took eighteen minutes to complete, was based on the double-antigen sandwich immunoassay principle. The three main benefits of the electrochemiluminescence immunoassay are increased specificity, a reduction in the number of reagents needed, and a shorter incubation time. A 20-microliter aliquot of the sample taken from the subjects is incubated with a mixture of biotinylated and ruthenylated RBD antigen as part of the assay's procedural execution. When corresponding antibodies are present, this results in the formation of double-antigen sandwich immune complexes. A microparticle coated with streptavidin is then added, which alters attachment to the previously mentioned complex. After that, the reagent mixture is put into a measuring cell, where an electrode captures the microparticles and simultaneously removes any unbound materials. Voltage is applied to precipitate electrochemiluminescence, which is then quantitatively measured using a photomultiplier. The resulting signal makes it easier to detect and measure IgG antibodies because it shows a positive correlation with increases in antibody titer values. The samples' pharmacological and medical anamneses were conclusively determined during the data collection phase. Individuals who had no prior medical or medication-related conditions before vaccination but who developed morbidities only after vaccination were more likely to attribute symptomatology to side effects caused by the COVID-19 Covishield vaccine. The Covishield vaccine was chosen as the main focus of this study because it is more widely used than other vaccination methods.

RESULT AND DISCUSSION

A cohort of 350 participants was randomly selected for this study and subsequently stratified based on their age. The demographic distribution indicated that the 21-30 age bracket received the majority of samples (Figure 1), while the 51-55 age group was the least represented.

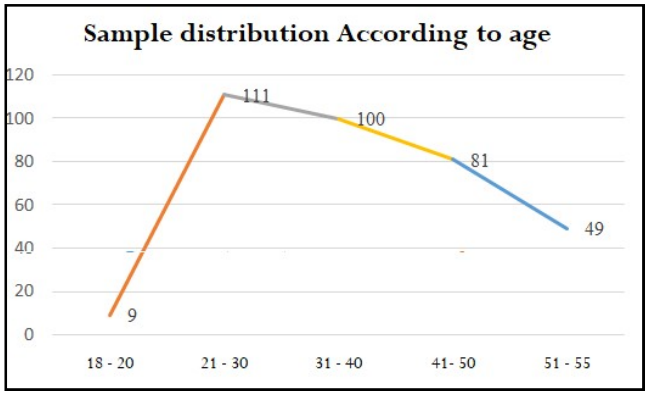

Figure 1.

This demographic is likely to provide valuable insights into the research question, which is why the study's relevance is enhanced by the predominance of samples from the 21-40 age range. A crucial discovery in this study relates to the seroconversion results, as a significant percentage (81%) of the sampled population demonstrated antibody positivity (Figure 2).

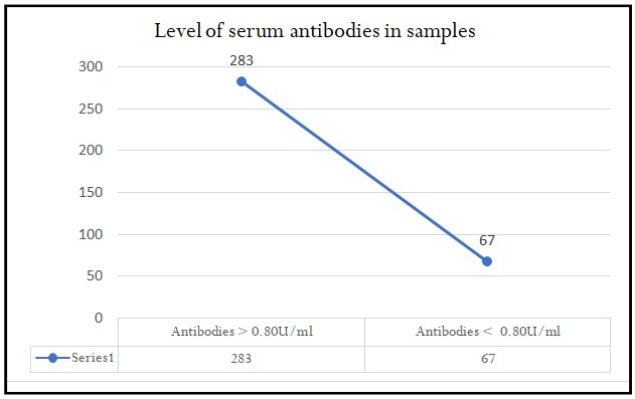

Figure 2.

Conversely, the remaining 19% of samples did not reach the necessary antibody threshold (>0.80 U/ml) required for sufficient immunity against COVID-19, according to established literature. This dichotomy in antibody response highlights the variability in individual immune reactions to vaccination. This graphical representation illustrates a comparative analysis of gender-specific differences in seroconversion efficacy regarding COVID-19. Despite an equitable distribution of participants by gender, a significant disparity in antibody development was observed between male and female groups. In the female subset of 175 samples, a significant number (47 samples) showed no seropositivity, while in the male subset of the same size, a lesser number (20 samples) exhibited a similar absence of antibody production (Figure 3). The findings indicate a gender-mediated limitation in the humoral immune response to COVID-19, with females seemingly more adversely impacted, demonstrating a reduced ability for antibody production compared to males. Significantly, among male samples, individuals aged 51-55 demonstrated the greatest prevalence of insufficient antibody response to COVID-19 (Figure 4). This observation indicates that age is a significant factor in seroconversion, with older adults likely encountering difficulties in generating an effective immune response. Moreover, the existence of comorbidities in this demographic may diminish vaccine efficacy, consequently impeding antibody production.

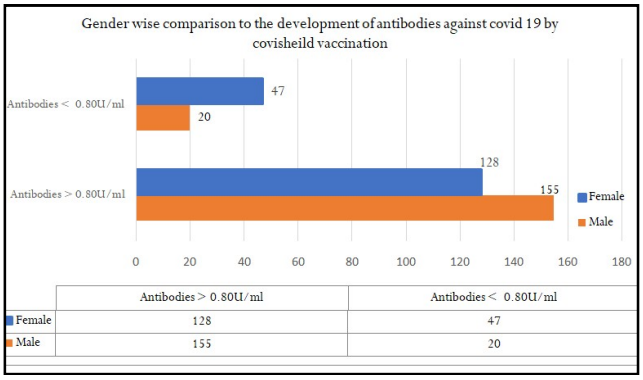

Figure 3.

The presence of multiple comorbidities in individuals within this age group may further aggravate this issue, ultimately hindering the development of a strong immune response. Clarifying the mechanisms that lead to inadequate antibody production is a critical research priority, as the specific immunological pathways involved remain unclear.

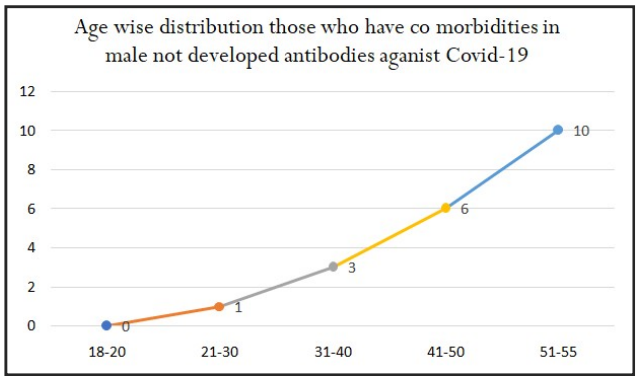

Figure 4.

The comorbidity profile indicated a predominance of diabetes mellitus, followed by hypertension (HTN), myocardial infarction (MI), asthma, and stroke, among others (Figure 5). The majority of individuals with these comorbidities were aged 51 to 55. The results indicate that adults without underlying comorbidities are likely to demonstrate a more advantageous response to pandemic vaccination, thus providing a relative safety benefit. This study recommends a prudent strategy for future pandemic situations, emphasizing that elderly individuals and those with lifestyle-related diseases should maintain increased vigilance and adopt additional precautions, even after vaccination, to reduce potential risks and enhance protection.

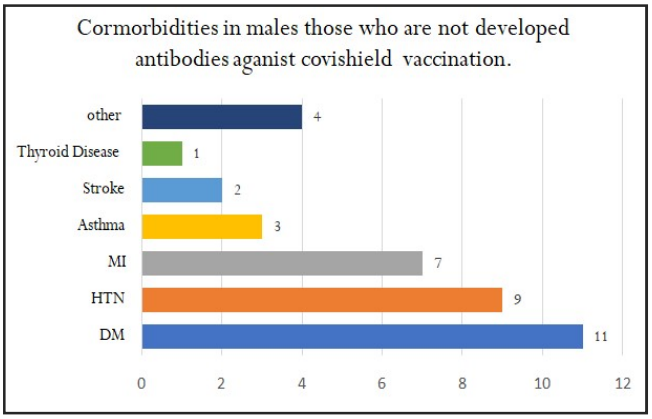

Figure 5.

The graphical representation illustrates the age-wise distribution of individuals who failed to develop antibodies against COVID-19. A comparative analysis of both genders reveals a preponderance of samples within the 41-55 age bracket(Figure 6). Notably, among males, the majority of samples emanated from the 51-55 age group, suggesting a potential correlation between advanced age and diminished seroconversion efficacy. This graphical representation illustrates a comparative analysis of males and females, categorized

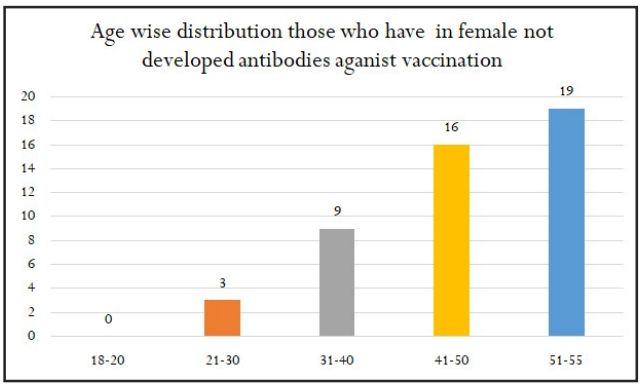

Figure 6.

by age, who demonstrated an absence of seroconversion to COVID-19. The data indicates a significant trend in which most female samples that did not produce antibodies were aged 41-55, while the primary age group for males was 51-55 (Figure 7). These findings highlight the importance of age and comorbidities as essential factors influencing antibody development, indicating that individuals in these age groups, especially those with preexisting health conditions, may be more vulnerable to compromised immune responses to the pandemic.

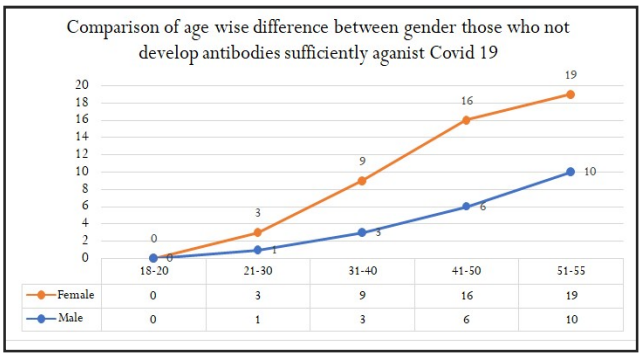

Figure 7.

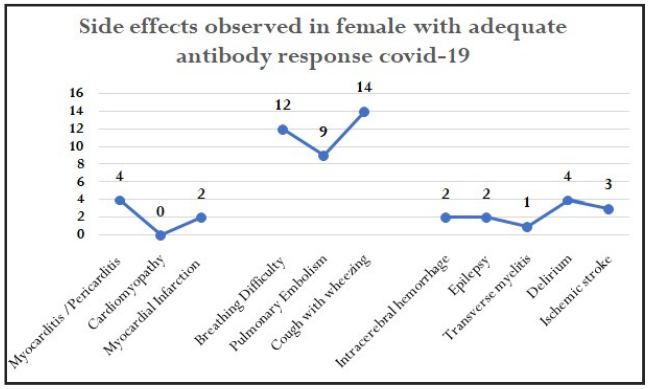

Figure 8.

The graphical representation illustrates the occurrence of side effects in female subjects who exhibited a strong immune response, as indicated by the production of adequate antibody titers following Covishield vaccination. Among 128 participants, a specific subset displayed a range of side effects, which were carefully recorded and temporally associated with vaccination.

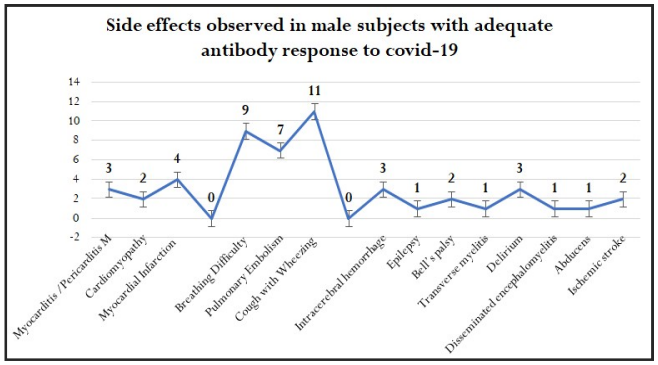

Figure 9.

These individuals lacked any prior medical or pharmacological history that could have potentially influenced the observed reactions. The temporal relationship between vaccination and the onset of side effects at various intervals strongly suggests that the vaccine is the likely causal agent. This study conducted a thorough examination of the side effects associated with Covishield vaccination, specifically targeting three primary physiological systems: cardiovascular, respiratory, and neurological. The neurological domain exhibited a marked inclination towards severe manifestations, encompassing ischemic stroke in four samples and delirium in three samples (figure 8). In contrast, the pulmonology cohort primarily exhibited respiratory symptoms, notably cough and wheezing. Four samples within the cardiovascular system exhibited myocarditis. The side effect profile indicates that Covishield vaccination may be linked to hemodynamic disturbances, potentially leading to circulatory dysfunction. A comparative analysis with male samples indicated a distinct side effect profile, with females demonstrating a comparatively reduced likelihood of neurological sequelae.

The graph depicts the occurrence of side effects in male individuals who achieved adequate antibody titers following the Covishield vaccination. Within a cohort of 155 samples, a subset of individuals exhibited side effects, which were recognized and recorded post-immunization. Significantly, these individuals possessed no antecedent medical or pharmacological history that could have obscured the observed reactions. The temporal correlation between vaccination and the onset of side effects at different intervals strongly indicates a causal relationship, implicating the vaccine as the likely source of the side effects. This study examined the adverse effects linked to the Covishield vaccine, focusing specifically on three critical physiological systems: cardiovascular, respiratory, and neurological(Figure 9). The neurological domain displayed the most severe manifestations, with three samples suffering from ischemic stroke and delirium. Conversely, the pulmonology cohort primarily exhibited respiratory symptoms, including cough and wheezing. Four samples experienced myocardial infarctions within the cardiovascular system. The side effect profile indicates that vaccination with Covishield may be linked to circulatory disturbances. Nonetheless, these findings should not be interpreted as a condemnation of the vaccine's safety or as an advisement against vaccination. Due to the critical significance of immunization in reducing pandemic-related illness and death, it is essential for researchers to carefully observe and analyze potential risks linked to vaccine administration, thus facilitating informed risk-benefit evaluations.

CONCLUSION

This study's findings clearly illustrate the essential role of vaccination in addressing pandemics, especially regarding COVID-19.

The rapid development of vaccines, including Covishield, has been crucial in enhancing immune responses and averting disease transmission. This investigation offers persuasive evidence regarding the efficacy and significance of vaccination in alleviating pandemics, notwithstanding the controversies and debates related to vaccine development. It is crucial to prioritize post-vaccination monitoring and adverse event surveillance to clarify the potential risks and benefits of vaccine administration. By doing so, healthcare professionals can guarantee the prudent application of vaccines and enhance patient safety.

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Conflict of interest: None

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Ethics statement: The human ethics committee approval has been obtained from JJTU.

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