



SARS Antibodies NCP, RBD and S2 in Women Vaccinated with Booster Pfizer Vaccine and Infected in Meantime with SARS-CoV-2

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Abstract

The effective immunization plays a key role in preventing the spread of SARS-CoV-2 infection. The case study describes the serum titer of antibodies against NPC, RBD and S2 SARS-CoV-2 in 69-year-old women, who developed hybrid immunity. She was vaccinated with booster dose of Pfizer vaccine and was twice infected with SARS-CoV-2. Despite vaccination and booster dose, she was re-infected. Five months after the booster dose of the vaccine, serum levels of antibodies to RBD (806 U/mL) and S2 – (452 U/ml) were detected.

Introduction

In November 2019, in the city of Wuhan, Hubei Province, China, the novel coronavirus was identified as the cause of pneumonia. The virus spread rapidly, resulting in an epidemic throughout China, followed by a global pandemic. This Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) appears to be a new human pathogen and The World Health Organization (WHO) defined the disease as COVID-19 in February 2020 [1-3]. According to The Chinese Center for Disease Control and Prevention 81% of COVID-19 cases are classified as mild (no pneumonia or mild pneumonia), 14% as severe, and 5% as critical [4]. The effective immunization plays a key role in preventing the spread of SARS-CoV-2 infection by evoking the production of virus-specific neutralizing antibodies and long-lived memory B cells [5,6]. Monitoring the immune response against SARS-CoV-2 is pivotal in the evaluation of long-term vaccine efficacy. The outer surface of SARS-CoV-2 contains the spike (S), matrix (M), and envelope (E) proteins. The S protein plays a role in viral host range and infectivity—it is a critical target for inducing antibodies, particularly Neutralizing Antibodies (NAbs) specific against SARS-CoV-2 [7].

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Case Presentation

The 69-year-old woman, not infected with SARS-CoV-2 since the beginning of the pandemic, was vaccinated in January 2021 against COVID-19 with BNT162b2 mRNA COVID-19 vaccine (Comirnaty; BNT162b2, BioNTech/Pfizer, Mainz, Germany/New York, NY, USA), next in March 2021. After nine months, in November 2021 she was vaccinated with booster dose. In meantime, before the booster vaccination she was mildly infected. Nasopharyngeal swabs were taken for SARS-CoV-2. RT-PCR test was positive. In April 2022 she was infected once again (antigenic test was weak positive); the symptoms were mild and she completely recovered without sequelae.

Serum specimens for antibody detection were collected:

- Two months after second dose
- 9 months after second dose
- Two weeks after infection and before booster vaccination
- Two months after booster vaccination
- Two weeks after the reinfection and 5 months after booster vaccination

Materials and Methods

The nasopharyngeal sample was previously extracted using an automated TANBead Maelstrom 8 (TANBead Nucleic Acid Extraction Kit), and tested for SARS-CoV-2 at the SARS Laboratory of the Medical University in Lublin, Poland. The genisig' Real-Time PCR Coronavirus



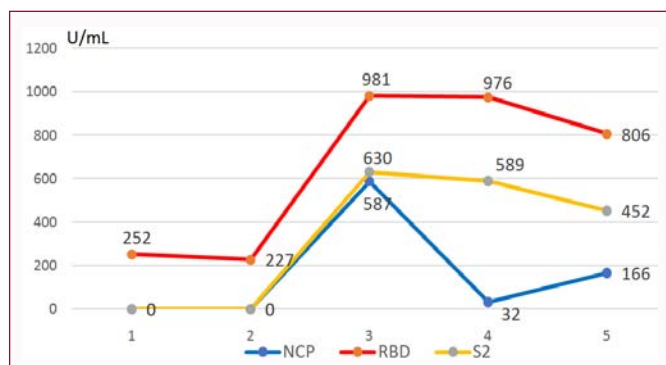


Figure 1: NCP, RBD and S2 IgG antibodies titer in the vaccinated and re-infected patient.

COVID-19 (CE IVD) was used to detect SARS-CoV-2 viral RNA (Primerdesign Ltd, School Lane, Chandler’s Ford, UK). Reaction system and amplification conditions were performed according to the manufacturer’s instruction. Serum was obtained from a 5 ml blood sample after centrifugation at 3600 rpm for 10 min at room temperature, then frozen and maintained at – 80°C till usage. Serum samples were thawed at 37°C for 10 min and after vortexing they were analyzed for the detection of IgG against SARS-CoV-2 through the Microblot-Array COVID-19 IgG, IgA and IgM (TestLine Clinical Diagnostics Ltd, Brno, Czech Republic – CoVMA96) according to the manufacturer’s instructions. Immunogenicity results are reported as an international standard unit (IU/mL). Briefly, the immunoassay evaluates antibodies against NPC, RBD and S2 antigen. The results are interpreted as follows: <185 U/mL = negative, 185–210 U/mL= borderline, >210 U/mL = positive. The research was approved by the Medical University of Lublin Ethics Committee and by GCP (Good Clinical Practice) regulations (No. KE-0254/295/2019, 26 September 2019). Written informed consent was obtained from the patient.

Results

Two months after two doses of vaccine the level of RBD antibody was 252 U/mL and 9 months later the antibody titer decreased – 227 U/mL. In meantime the patient was infected and two weeks after infection and before booster vaccination in serum was detected high level of RBD antibody – 981 U/mL, nucleocapsid protein antibody (NCP) - 587 U/mL and antibody against S2 – 587 U/mL. Two months after booster dose RBD - 976 U/mL and S2 - 589 U/mL but NCP was undetectable. Five months after booster vaccination the patient was re-infected and two weeks later in the serum was present only antibody against RBD – 806 U/mL and S2 – 452 U/ml. NCP antibody was undetectable. Despite vaccination, the patient was infected twice. RBD and S2 antibody titer declined. Moreover, the reinfection did not increase the antibody titer (Figure 1).

Discussion

In the natural infection by SARS-CoV-2 the major antigenic target of human antibodies is the Spike protein, especially the Receptor Binding Domain (RBD), which is responsible for the binding to the Angiotensin-Converting Enzyme 2 (ACE2), the main receptor recognized by the virus to enter into the target cells, and is the primary target of SARS-CoV-2-neutralizing antibodies [8,9]. Antibody response against this protein and, in particular, the RBD represents a valuable tool to monitor the vaccine efficacy [10]. Dorigatti et al. [11] suggest that SARS IgG antibody correlate with age, and with a previous history of COVID-19, and mRNA vaccination.

A majority of people infected with SARS-CoV-2 will recover from the primary infection. Some of them are re-infected. Kim et al. [12] described a close correlation between neutralizing antibody titer and SARS-CoV-2 reinfection. Primary infection-induced high NAb titers may not completely protect the host from reinfection. In order to hamper the continuous spread of this virus, ideally recovered patients would have sufficient neutralizing antibodies to protect themselves against reinfection. Our patient had reinfection 5 months after the booster dose of vaccine. Study performed by Giorgi [13] suggest that immunological memory is acquired in most individuals infected with SARS-CoV-2 and is sustained in a majority of patients for up to 11 months after recovery. Numerous of studies have reported that asymptomatic COVID-19 patients exhibit lower antibody responses than patients with severe COVID-19. Moreover, there is rapid decline of anti-SARS-CoV-2 antibody responses in asymptomatic COVID-19 patients compared with severe COVID-19 patients [14-16]. Thus, asymptomatic patients may be at high risk of reinfection. The described patient developed weak and unspecific COVID-19 symptoms both in first infection and re-infection. She developed a hybrid immunity (SARS-CoV-2 infection and vaccination against SARS-CoV-2) which appears to be most protective. Protection with one or two doses of vaccine following natural infection was significantly greater than protection associated with natural infection alone. Asymptomatic infection is associated with lower virus-specific IgG antibodies concentrations compared to those with symptomatic infections [17]. Five months after the booster dose of the vaccine and additionally two mild COVID diseases, in the serum of described patient only antibodies against S2 protein and RBD were detected. The level of the NCP antibody that had been detected 5 months earlier had dropped below the threshold of detection.

Conclusion

Despite vaccination and booster dose, mild reinfection is possible. Five months after the booster dose of the vaccine, serum levels of antibodies to RBD (806 U/mL) and S2 – (452 U/ml) were detected.

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