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Antibody Production after COVID-19 Vaccination in Patients with Inborn Errors of Immunity

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ABSTRACT

Background: Few studies have evaluated COVID-19 vaccine efficacy in patients with inborn errors of immunity (IEI).

Objective: To evaluate the levels of antibody (Ab) production and function after COVID-19 vaccination in IEI patients with phagocytic, complement, and Ab deficiencies and their comparison with healthy controls.

Methods: Serum samples were collected from 41 patients and 32 healthy controls at least one month after the second dose of vaccination, while clinical evaluations continued until the end of the third dose. Levels of specific anti-receptor-binding domain (RBD) IgG and anti-RBD neutralizing antibodies were measured using EUROIMMUN and ChemoBind kits, respectively. Conventional SARS-CoV-2 neutralization test (cVNT) was also performed. Cutoff values of ≤20, 20-80, and ≥80 (for cVNT and Chemobined) and 0.8-4.2, 4.2-8.5, and ≥8.5 (for EUROIMMUN) were defined as negative/weak, positive/moderate, and positive/significant, respectively.

Results: A considerable distinction was observed between the Ab-deficient patients and the controls for Ab concentration (EUROIMMUN, $p<0.01$) and neutralization (ChemoBind, $p<0.001$). However, there was no significant difference compared with the other patient groups. A near-zero cVNT in Ab-deficient patients was found compared to the controls ($p<0.01$). A significant correlation between the two kits was found using the whole data ($R^2=0.82$, $p<0.0001$).

Conclusion: Despite varying degrees of Ab production, all Ab deficient patients, as well as almost half of those with complement and phagocytic defects, did not effectively neutralize the virus (cVNT). In light of the decreased production and efficiency of the vaccine, a revised immunization plan may be needed in IEI.

Keywords: COVID-19 Vaccines, Immunologic Deficiency Syndromes, Neutralization Tests, Viral Antibodies

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INTRODUCTION

Since the discovery of Coronavirus disease 2019 (COVID-19) in December 2019, the virus has impacted almost all patient groups throughout the world. However, some groups of people, such as those with inborn errors of immunity (IEI), may have weaker immune reactions to vaccines (1). IEI is a heterogeneous group of disorders characterized by defects in innate or adaptive immunity, leading to increased susceptibility to infections and other complications (2). IEI includes over 300 distinct illnesses that impact different parts of the immune system, including B cells, T cells, natural killer cells, phagocytes, complement system, and cytokines (3). Depending on the type and severity of IEI, patients may suffer from recurrent or severe infections, autoimmunity, allergy, inflammation, malignancy, or organ dysfunction (4).

Since both the innate and adaptive immune responses are essential in deterring the virus, patients with various types of immunodeficiencies are afflicted (5). A mass vaccination policy is a reasonable approach to counteracting the effects of the disease on the general population and IEI patients (6). Nevertheless, there is insufficient conclusive data regarding the effects of COVID-19 and its vaccination on patients with IEI. To guide clinical practice and public health policies, additional data is necessary (7).

The impact of COVID-19 on IEI patients is not fully understood. On the one hand, IEI patients may be more vulnerable to SARS-CoV-2 infection and its complications due to their impaired immune function and comorbidities (8-11). On the other hand, some IEI patients may have a less severe course of COVID-19 due to their impaired inflammatory response or existing immunity, or the type of treatment protocols like using intravenous immunoglobulin (IVIG) to protect themselves (10, 12). Therefore, it is important to monitor the clinical outcome and immune response of IEI patients after COVID-19 infection or vaccination. Several studies have reported the clinical characteristics and outcomes of COVID-19 in IEI patients. Drzymalla et al. conducted a systematic review comprising 68 articles, involving 459 patients with IEI who were diagnosed with COVID-19 (1). The authors calculated a case fatality rate of 9%, a hospitalization rate of 49%, and an oxygen supplementation rate of 29%. The authors found that patients with antibody production defects had worse outcomes than the other groups. However, they also noted that the data were limited by heterogeneity, selection bias, and reporting bias.

The effectiveness and safety of COVID-19 vaccines in IEI patients are also being studied (13, 14). Several studies have shown that certain patients with IEI exhibited various types of immune response after receiving the COVID-19 vaccination (15). The aim of this study was to evaluate antibody production after the second dose of COVID-19 vaccination and clinical evaluations for COVID-19 following every dose of vaccination in IEI patients and compare it with healthy controls.

MATERIALS AND METHODS

Study Design and Sample Size

This cross-sectional study included 41 IEI patients and 32 healthy controls. The inclusion criteria for the IEI patients were: 1) having a confirmed diagnosis of IEI by a clinical immunologist before the study; 2) receiving at least one dose of COVID-19 vaccine; and 3) providing informed consent. The exclusion criteria for the IEI patients were: 1) having a history of blood transfusion or immunoglobulin therapy within 4 to 6 weeks before the study, and 2) having any other chronic or acute diseases that could affect the immune response. The diagnosis of IEI had been established before the investigation by a clinical immunologist at Immunology, Asthma, and Allergy Research Institute (IAARI), Tehran, Iran. IEI diagnosis for all

patients was made using clinical evaluation, and immunological assessments including tetrazolium test (NBT), dihydrorhodamine (DHR), immunoglobulins, antibody titration, complement assessment, lymphocyte transformation test (LTT), and genetic analysis. This study has been approved by the ethics committee of Immunology, Asthma and Allergy research Institute (IAARI) with the approval code: IR.TUMS.IAARI.REC.1399.005.

Data Collection Procedures

Blood samples were obtained from both patients and controls after the second dose of the COVID-19 vaccine and stored at -20 °C for later analysis. Participants were also surveyed using a questionnaire, gathering information such as demographic details, the specific type of immune system disorder (for IEI patients), the brand of vaccine received (Sinopharm, AstraZeneca, or Sputnik), the number of vaccine doses administered, and any COVID-19-like symptoms experienced before and after each vaccination dose. Blood samples from all participants were tested at least one month after the second vaccination. Data for the study was collected through a telephone survey and blood samples obtained between October 2020 and July 2021.

Conventional Virus Neutralization Test (cVNT)

A conventional virus neutralization test (cVNT) was performed with the "Wuhan" SARS-CoV-2 strain, isolated using a nasal swab from an Iranian patient and characterized after several passages on VeroE6 cells (as target cells). Using the Spearman-Karber method, the amount of SARS-CoV-2 virus can be determined by a 50% tissue culture infectious dose (TCID50) endpoint dilution assay. This assay requires infecting cell cultures with a fixed amount of virus and different amounts of serum having neutralizing antibodies against SARS-CoV-2, and then checking the cytopathic effect (CPE) of the virus on the cells. The end-point is the highest serum dilution that blocks at least 50% of CPE in the cell cultures. The virus titer is then computed using a formula that considers the dilution factor, the number of wells tested, and the fraction of wells with CPE. The virus titer can be shown as TCID50 per ml or TCID50 per gram of tissue (16).

In this study, a two-fold serial dilution of serum samples was done in a 96-well microplate, from 1:2 to 1:1024 (1:210) in triplicates. The dilutions were mixed with an equal volume of SARS-CoV-2 (100 TCID50/mL) and then transferred to wells containing monolayers of target cells. The microplate was sealed with pressure film and incubated at 4 °C. After 60 minutes, it was transferred to 37 °C in a 5% CO^2 incubator and monitored daily for the viral cytopathic effect not inhibited by the neutralization antibody. Each microplate assay included 1) blank control wells for cell culture (serum-free and virus-free), 2) positive standard serum samples (30 serum samples were collected from patients with a history of COVID-19, tested and confirmed with a specific titer, then pooled), 3) negative control serum samples containing a pool of serum samples collected 15 years before the COVID-19 pandemic (2004), and 4) the positive control for the virus (serum-free wells containing isolated and characterized SARS-CoV-2 with a virus titration of 30 to 300 $\log_{10}$ $CCID_{50}$/mL). In each microplate, 4 wells were allocated for each of the four above-mentioned control samples. After 4 to 6 days of incubation at 37 °C, the virus-neutralizing antibody titer at 50% inhibition was calculated by the Spearman-Karber method (17). Cutoff values of ≤20, 20-80 and ≥80 were defined as negative/weak, positive/moderate, and positive/significant, respectively.

Specific Anti-RBD IgG using EUROIMMUN ELISA Kit

The coated microplates from EUROIMMUN KIT (EI 2606-9601 G) were used for evaluating specific IgG against SARS-CoV-2 using enzyme-linked immunosorbent

assay (ELISA). The ready-to-use plates were coated with the immunologically relevant receptor binding domain (RBD) of the S1 subunit of the spike protein and the test was done according to the manufacturer's instructions. To estimate the samples in a semi-quantitative way, a ratio was computed from the extinction of the sample and that of the calibrator. Subsequently, the samples were categorized using modified cutoffs derived from Iranian healthy control data. Cutoff values of 0.8−4.2, 4.2−8.5, and ≥8.5 were considered negative/weak, positive/moderate, and positive/significant, respectively.

Anti-RBD Neutralizing Antibody Test

The conventional virus neutralization test is the preferred way to detect functional, SARS-CoV-2-specific neutralizing antibodies in serum samples. Another option is surrogate ELISA using ACE2 as the target structure to detect antibodies neutralizing the virus (18).

Neutralizing antibodies against the viral RBD were quantitatively assessed using a commercial ELISA kit (ChemoBind, Tehran, Iran) according to the manufacturer's instructions. Initially, 100 µL of 6 dilutions of standards: 0, 10, 20, 40, 80, and 120 were added to each well. The negative and positive controls (provided by the kit) were seeded serially in each well already coated with RBD antigens. Then, the samples were diluted 100 times (5 µL serum plus 450 µL sample diluent), and 100 µL of each sample was added to each well. After 1 h of incubation, the plate was washed 3 times with a wash buffer, and 100 µL of Streptavidin anti-IgG antibody was added to each well, and incubated for 30 min.. After 3 times washing, 100 µL TMB (3,3',5,5'-tetramethylbenzidine) substrate was added to each well and the reaction was halted using the stop solution. The plate was read by an ELISA reader at 450 nm and 630 nm and concentrations were calculated using the standard curve. Cutoff values of ≤20, 20−100 and ≥100 were defined as negative/weak, positive/moderate, and positive/significant, respectively.

Statistical Analysis

The data analysis was performed using R software version 4.2. To harmonize the data and enable comparison across different units, the data from three methods were transformed using the inverse hyperbolic sine function. The Pearson correlation coefficient and its *p*-value were calculated using the `stat_cor` function in R, which uses a nonparametric bootstrap method to estimate the confidence interval of the coefficient. We employed the Kruskal-Wallis test to compare the groups, given that not all the groups exhibited a normal distribution. A *p*-value less than 0.05 was considered significant.

RESULTS

Patients' Characteristics

The participants comprised 27 men and 14 women in the IEI group, 27 females and 5 males in the control group. The median ages (min-max) during the sampling for the IEI group and the control group were 33 years (19–78) and 41 years (29–70), respectively. The IEI cases included patients with complement deficiency (hereditary angioedema: 19 cases), Ab deficiency [common variable immunodeficiency (CVID): 8 cases, hyper-IgE syndrome (HIES): 2 cases, and X-linked agammaglobulinemia (XLA): 2 cases] phagocytic function deficiency [chronic granulomatous disease (CGD): 7 cases and neutropenia: 3 cases]. Of the 41 patients, 39 (95%) had received Sinopharm vaccines and 2 (5%) had received AstraZeneca vaccines for all three doses. The distribution of vaccine types among the controls was as follows: AstraZeneca (n=29; 90.6%), Sinopharm (n=1; 6.3%), and Sputnik (n=2; 3.1%). The patient characteristics are shown in Table 1.

COVID-19 Before and After Vaccination among the Patients

Prior to the availability of COVID-19 vaccination, we conducted a phone-based survey as the initial phase of our project.

Table 1. Patients' characteristics

Patients	Diagnosis	Number of Patients	Gender (F/M)	Age (Years)
Complement Deficiency	HAE	19 (44.2%)	9/10	42.2±3.9*
Phagocytic Deficiency	Neutropenia	3 (7.3%)	1/2	27.3±1.8
	CGD	7 (17.1%)	2/5	32.1±2.7
Antibody Deficiency	XLA	2 (4.9%)	0/2	25.5±2.5
	CVID	8 (19.5%)	1/7	33.2±3.9
	HIES	2 (4.9%)	1/1	37.5±17.5

HAE, hereditary angioedema; CGD, chronic granulomatous disease; XLA, X-linked agammaglobulinemia; CVID, common variable immunodeficiency; HIES, hyper-immunoglobulin-E syndrome; *mean±SD

Table 2. Data on COVID-19 before and after three doses of vaccination

Patients	Covid-19 before vaccination	Covid-19 after 1st vaccination	Covid-19 after 2nd vaccination	Covid-19 after 3rd vaccination
HAE	8/19	7/19	3/19	1/19
Neutropenia	0/3	0/3	1/3	1/3
CGD	3/7	3/7	2/7	2/7
CVID	6/8	4/8	3/8	4/8
XLA	0/2	0/2	0/2	1/2
HIGE	1/2	1/2	1/2	1/2
Total	18/41	15/41	10/41	10/41

HAE, hereditary angioedema; CGD, chronic granulomatous disease; CVID, common variable immunodeficiency; XLA, X-linked agammaglobulinemia; HIES, hyper-immunoglobulin-E syndrome

The aim was to determine the prevalence of COVID-19 among patients with inborn errors of immunity (IEI) and assess the awareness of their families regarding adherence to the health protocols. According to our records, 44% (18/41), 36.6% (15/41), 24.4% (10/41), and 24.4% (10/41) of the patients reported a history of COVID-19-like symptoms before and after the first, second and third doses of vaccination, respectively (Table 2). Some patients suffered from the disease 2 or more times. Only one patient with HAE died due to an angioedema attack after the first dose of vaccination, and was therefore excluded from the study. Only two patients with CVID (Ab deficiency group) had a history of hospitalization due to hypoxemia (oxygen saturation less than 95%) and ground glass opacities (GGO) in their CT findings.

Antibody Production After COVID-19 Vaccination

The results of different antibody detection tests were grouped into 3 categories based on the increase in antibody function or levels: 'negative/weak', 'positive/moderate', and 'positive/significant'. The cutoff values for the anti-SARS-CoV-2 IgG EUROIMMUN ELISA Kit were modified based on the healthy controls (Iranian blood samples following exposure to the wild-type SARS-CoV-2 virus [Wuhan]). Cutoff points for ChemoBind were set according to the kit's instructions. Cutoff values of ≤20, 20−80, and ≥80 (for cVNT and ChemoBind kit) and 0.8−4.2, 4.2−8.5, and ≥8.5 (For EUROIMMUN ELISA kit) were defined as negative/weak, positive/moderate, and positive/significant, respectively.

As shown in Fig. 1, the descriptive analysis of the IEI patients in each subgroup (complement deficiency, phagocytic deficiency and antibody deficiency) has been illustrated following the second dose of vaccination compared with the healthy controls.

The ChemoBind anti-RBD neutralizing Ab test (Fig. 1A) and the EUROIMMUNE kit for specific anti-RBD IgG (Fig. 1B) indicated

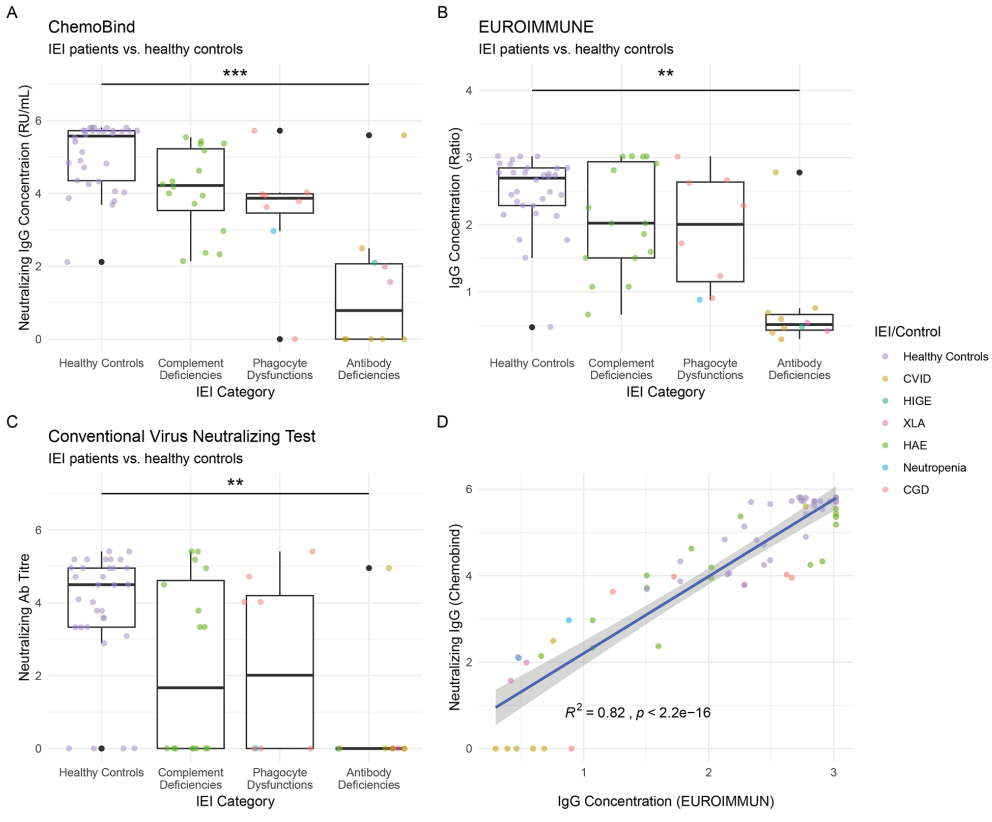

Fig. 1. Descriptive analysis of the patients in each IEI group (complement deficiency, phagocytic deficiency and antibody deficiency) and the healthy controls following the second dose of vaccination. The assessment of the anti-RBD neutralizing Ab test using the Chemobind kit (A), specific anti-RBD IgG using the EUROIMMUNE kit (B), and cVNT (C) for all the patients (n=41) and the healthy controls (n=32) revealed a notable disparity between antibody deficient patients and the healthy individuals, although a partial level of Ab concentration and function were found using the kits. Moreover, a correlation between the two methods of anti-RBD neutralizing Ab test by ChemoBind kit and IgG concentration using Euroimmune kit was found using the data of all the patients and the healthy controls (D). *p<0.05, **p<0.01, ***p<0.001. cVNT: conventional virus neutralization test

that individuals lacking complement and phagocyte function exhibited a wide range of Ab production slightly lower than in the healthy individuals. However, these differences were not statistically significant and there was no considerable difference between the two groups of phagocytic and complement patients. On the other hand, patients with antibody deficiencies had lower levels of antibodies using ChemoBind and EUROIMMUNE kits than both the complement and phagocytic deficient patients (not statistically significant) and the healthy controls (with a significance level of p<0.001 and p<0.01 for the ChemoBind and EUROIMMUNE kits, respectively).

While patients with impaired antibody production exhibited a diverse range of antibody production and function when assessed using the ChemoBind and EUROIMMUNE kits, the cVNT method (the gold standard test for neutralization) revealed a nearly negligible capacity for neutralization in these patients compared with the healthy controls. This finding, as illustrated in Fig. 1C, was notably different from the results observed in the healthy controls (p<0.001). As shown in Fig. 1D, a positive and strong correlation was found between anti-RBD neutralizing Ab test by ChemoBind kit and IgG concentration using the EUROIMMUNE kit by the linear regression model (R^2=0.82, p<0.0001). This model was calculated using cumulative data of patients (n=41) and the healthy controls (n=32).

DISCUSSION

Herein we conducted a nine-month study with two aims: first, to evaluate the clinical outcomes of IEI patients who might have been infected by COVID-19 before or after receiving the COVID-19 vaccine, using a phone survey; and second, to analyze the production and function of anti-SARS-CoV-2 antibodies after the second dose of vaccination.

Few studies have looked at how COVID-19 affects IEI patients and their antibody reactions (19). According to our data, prior to vaccination, 18 out of 41 patients (43%) exhibited symptoms of COVID-19. After receiving the COVID-19 vaccination, 36.6% experienced symptoms following the first dose, while 24.4% experienced symptoms after the second and third doses. Among the patients, two individuals were hospitalized due to the severity of their condition. In line with our study, Pieniawska-Śmiech et al. indicated that among 99 children with IEI, 26.5% got infected with SARS-CoV-2, only three needed hospital care and none died. After a year follow-up, 47.06% of them had positive tests for the anti-SARS-CoV-2 antibody and they mostly had mild and short-lived symptoms even among the patients with defects in IgG production (20, 21). The mortality rates of IEI patients with COVID-19 are higher, according to some reports, but others disagree on how often they get infected (8). In addition, an international study on 94 IEI patients concluded that COVID-19 risk factors in IEI patients are similar to those reported in the general population (22). Lower hospitalization rates and a general protective role for COVID-19 have also been described in IEI patients, particularly in those with B-cell dysfunction (5, 12, 23).

COVID-19 vaccine efficacy in IEI patients is also an area of investigation. Our findings revealed that patients with complement and phagocyte deficiencies displayed a wide range of antibody production, slightly lower than that of the healthy controls. However, these differences did not reach statistical significance. Interestingly, there were no significant differences observed between the two groups of phagocytic and complement patients, indicating that the impact of complement and phagocyte function on antibody production may be comparable. On the other hand to statistically significant" to "In contrast, individuals with antibody deficiencies showed notably reduced antibody levels, as indicated by both kits and the cVNT, in comparison to both complement and phagocytic deficient patients (though not reaching statistical significance) and healthy controls (which was statistically significant). These findings highlight the importance of antibody deficiencies in compromising the production and functionality of anti-SARS-CoV-2 antibodies, suggesting that individuals with antibody complications may have reduced protection against COVID-19. Further investigations are warranted to better understand the underlying mechanisms and explore potential interventions to enhance the immune response in these individuals. The absence of neutralizing antibodies in the cVNT test suggests that the limited amounts of antibodies produced in these patients, although not completely absent, lacked the potency to effectively counteract viral propagation.

In a survey by Hagin et al., specific IgG antibodies were detected after vaccination in 18 of 26 IEI patients, and 19 patients also developed an S-peptide–specific T cell response. Nevertheless, the antibody titer was lower than the level in the healthy population which was in accordance with our results (12, 21). This phenomenon is unlike what is seen in secondary immunodeficiencies (24-26). Although inactivated vaccines are regarded as safe, more evidence is needed to establish effective guidelines on the type and schedule of vaccines in different IEI subgroups (27).

Based on the impaired generation and function of anti-SARS-Cov2 antibodies in patients with antibody complications, different vaccination doses may not provide a protective effect, which needs to be

elucidated. We did not measure the valency of the spike protein or the antibodies, as this was beyond the scope of our study. However, we acknowledge that valency may play a role in the diverse antibody responses observed in the IEI patients and the healthy controls.

In addition, our results showed a strong positive correlation between the IgG concentration measured using the EUROIMMUN kit and the neutralizing capacity assessed by the ChemoBind kit, indicating a close link between antibody production and functional effectiveness in our study population. This significant correlation emphasizes the importance of considering both aspects when assessing the overall effectiveness of the immune system against the target antigen. Similar findings were reported by Dolscheid-Pommerich et al., who investigated the association between a quantitative ELISA (IgG) and a microneutralization assay in COVID-19 outbreak study populations (28).

The clinical implications of this study are that COVID-19 vaccination may induce antibody production in most IEI patients, except for those with antibody production defects. However, antibody production alone may not be sufficient to protect against COVID-19 infection and its complications. Therefore, IEI patients should continue to follow preventive measures such as wearing masks and social distancing. Moreover, IEI patients may benefit from other interventions such as immunoglobulin therapy or monoclonal antibodies (29).

This study has some limitations that need to be acknowledged: 1) the sample size was small and unbalanced between the IEI patients and the controls; 2) the IEI patients were heterogeneous and included different types of immunodeficiencies that may have different responses to COVID-19 infection and vaccination; 3) the controls were not matched with the IEI patients in terms of age, gender, vaccine type, or COVID-19 history; 4) the age range does not include the pediatric population and does not represent the whole population of the IEI patients. Moving forward, further research is warranted to conduct larger and more rigorous studies that compare antibody production after COVID-19 vaccination in the IEI patients using different methods. Additionally, investigating the correlation between antibody production and clinical outcomes is crucial. Exploring other aspects of the immune response, such as cellular immunity and cytokine levels, could provide valuable insights into COVID-19 infection and vaccination in the IEI patients (30).

CONCLUSION

In conclusion, while IEI patients are generally considered to be at higher risk for severe COVID-19 due to their impaired immune function and comorbidities, our survey suggests that some IEI patients may experience a milder course of the disease. As a result of our study, all IEI patients had weaker Ab production and function compared with the healthy controls, but this difference was much bigger in patients with problems in making antibodies. These findings emphasize the importance of monitoring the immune response and clinical outcomes of the IEI patients after the vaccination. While vaccination is a crucial strategy to combat the effects of the disease, it may be necessary to explore alternative approaches.

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AUTHORS' CONTRIBUTION

The authors confirm contributions to the

article as follows: work conception: MN; study design: MN and ZP; clinical diagnosis of the patients: MF and LM; performing the test and interpretation of data: EF and AM; analysis of the results: MM; interpretation of the results: MF and LM; draft manuscript preparation: MN; revising the text: MM and AM. All authors reviewed the final version and approved the paper to be published.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

REFERENCES

1. Drzymalla E, Green RF, Knuth M, Khoury MJ, Dotson WD, Gundlapalli A. COVID-19-related health outcomes in people with primary immunodeficiency: A systematic review. Clin Immunol. 2022;243:109097.
2. Picard C, Al-Herz W, Bousfiha A, Casanova JL, Chatila T, Conley ME, et al. Primary Immunodeficiency Diseases: an Update on the Classification from the International Union of Immunological Societies Expert Committee for Primary Immunodeficiency 2015. J Clin Immunol. 2015;35(8):696-726.
3. Bousfiha A, Jeddane L, Picard C, Ailal F, Bobby Gaspar H, Al-Herz W, et al. The 2017 IUIS Phenotypic Classification for Primary Immunodeficiencies. J Clin Immunol. 2018;38(1):129-43.
4. Cordero E, Goycochea-Valdivia W, Mendez-Echevarria A, Allende LM, Alsina L, Bravo García-Morato M, et al. Executive Summary of the Consensus Document on the Diagnosis and Management of Patients with Primary Immunodeficiencies. Enferm Infecc Microbiol Clin (Engl Ed). 2020;38(9):438-43.
5. Al Yazidi LS, Al Rawahi H, Al Busaidi I, Al Tamemi S. COVID-19 AND PRIMARY IMMUNODEFICIENCY: ONE-YEAR EXPERIENCE. J Paediatr Child Health. 2021;57(4):594.
6. Gathmann B, Grimbacher B, Beauté J, Dudoit Y, Mahlaoui N, Fischer A, et al. The European internet-based patient and research database for primary immunodeficiencies: results 2006-2008. Clin Exp Immunol. 2009;157 Suppl 1(Suppl 1):3-11.
7. Antonelli M, Penfold RS, Merino J, Sudre CH, Molteni E, Berry S, et al. Risk factors and disease profile of post-vaccination SARS-CoV-2 infection in UK users of the COVID Symptom Study app: a prospective, community-based, nested, case-control study. Lancet Infect Dis. 2022;22(1):43-55.
8. Delavari S, Abolhassani H, Abolnezhadian F, Babaha F, Iranparast S, Ahanchian H, et al. Impact of SARS-CoV-2 Pandemic on Patients with Primary Immunodeficiency. J Clin Immunol. 2021;41(2):345-55.
9. Esenboga S, Ocak M, Akarsu A, Bildik HN, Cagdas D, Iskit AT, et al. COVID-19 in Patients with Primary Immunodeficiency. J Clin Immunol. 2021;41(7):1515-22.
10. Marcus N, Frizinsky S, Hagin D, Ovadia A, Hanna S, Farkash M, et al. Minor Clinical Impact of COVID-19 Pandemic on Patients With Primary Immunodeficiency in Israel. Front Immunol. 2020;11:614086.
11. Milota T, Smetanova J, Bartunkova J. Clinical Outcome of Coronavirus Disease 2019 in Patients with Primary Antibody Deficiencies. Pathogens. 2023;12(1).
12. Soresina A, Moratto D, Chiarini M, Paolillo C, Baresi G, Focà E, et al. Two X-linked agammaglobulinemia patients develop pneumonia as COVID-19 manifestation but recover. Pediatr Allergy Immunol. 2020;31(5):565-9.
13. Milito C, Cinetto F, Garzi G, Palladino A, Puca M, Brambilla E, et al. Safety of mRNA COVID-19 Vaccines in Patients with Inborn Errors of Immunity: an Italian Multicentric Study. J Clin Immunol. 2023;43(2):299-307.
14. Parente R, Sartorio S, Brussino L, De Pasquale T, Zoli A, Agolini S, et al. Multicentric Observational Study on Safety and Tolerability of COVID-19 Vaccines in Patients with Angioedema with C1 Inhibitor Deficiency: Data from Italian Network on Hereditary and Acquired Angioedema (ITACA). Vaccines [Internet]. 2023; 11(4).
15. Erra L, Uriarte I, Colado A, Paolini MV, Seminario G, Fernández JB, et al. COVID-19 Vaccination Responses with Different Vaccine Platforms in Patients with Inborn Errors of Immunity. J Clin Immunol. 2023;43(2):271-85.
16. Matusali G, Colavita F, Lapa D, Meschi S, Bordi L, Piselli P, et al. SARS-CoV-2 Serum Neutralization Assay: A Traditional Tool for a Brand-New Virus. Viruses. 2021;13(4).
17. Ramakrishnan MA. Determination of 50% endpoint titer using a simple formula. World J Virol. 2016;5(2):85-6.
18. Sholukh AM, Fiore-Gartland A, Ford ES, Miner MD, Hou YJ, Tse LV, et al. Evaluation of Cell-Based and Surrogate SARS-CoV-2 Neutralization Assays. J Clin Microbiol. 2021;59(10):e0052721.
19. Milota T, Strizova Z, Smetanova J, Sediva A. An

immunologist's perspective on anti-COVID-19 vaccines. Curr Opin Allergy Clin Immunol. 2021;21(6):545-52.
20. Pieniawska-Śmiech K, Kuraszewicz A, Sado J, Śmiech K, Lewandowicz-Uszyńska A. Assessment of COVID-19 Incidence and the Ability to Synthesise Anti-SARS-CoV-2 Antibodies of Paediatric Patients with Primary Immunodeficiency. J Clin Med. 2021;10(21).
21. Hagin D, Freund T, Navon M, Halperin T, Adir D, Marom R, et al. Immunogenicity of Pfizer-BioNTech COVID-19 vaccine in patients with inborn errors of immunity. J Allergy Clin Immunol. 2021;148(3):739-49.
22. Meyts I, Bucciol G, Quinti I, Neven B, Fischer A, Seoane E, et al. Coronavirus disease 2019 in patients with inborn errors of immunity: An international study. J Allergy Clin Immunol. 2021;147(2):520-31.
23. Quinti I, Lougaris V, Milito C, Cinetto F, Pecoraro A, Mezzaroma I, et al. A possible role for B cells in COVID-19? Lesson from patients with agammaglobulinemia. J Allergy Clin Immunol. 2020;146(1):211-3.e4.
24. Boyarsky BJ, Werbel WA, Avery RK, Tobian AAR, Massie AB, Segev DL, et al. Antibody Response to 2-Dose SARS-CoV-2 mRNA Vaccine Series in Solid Organ Transplant Recipients. Jama. 2021;325(21):2204-6.
25. Peled Y, Ram E, Lavee J, Sternik L, Segev A, Wieder-Finesod A, et al. BNT162b2 vaccination in heart transplant recipients: Clinical experience and antibody response. J Heart Lung Transplant. 2021;40(8):759-62.
26. Bonelli MM, Mrak D, Perkmann T, Haslacher H, Aletaha D. SARS-CoV-2 vaccination in rituximab-treated patients: evidence for impaired humoral but inducible cellular immune response. Ann Rheum Dis. 2021;80(10):1355-6.
27. Bonilla FA. Update: Vaccines in primary immunodeficiency. J Allergy Clin Immunol. 2018;141(2):474-81.
28. Dolscheid-Pommerich R, Bartok E, Renn M, Kümmerer BM, Schulte B, Schmithausen RM, et al. Correlation between a quantitative anti-SARS-CoV-2 IgG ELISA and neutralization activity. J Med Virol. 2022;94(1):388-92.
29. Wolf J, Abzug MJ, Anosike BI, Vora SB, Waghmare A, Sue PK, et al. Updated Guidance on Use and Prioritization of Monoclonal Antibody Therapy for Treatment of COVID-19 in Adolescents. J Pediatric Infect Dis Soc. 2022;11(5):177-85.
30. Moss P. The T cell immune response against SARS-CoV-2. Nat Immunol. 2022;23(2):186-93.