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Cellular Immunity in Individuals Classified by Antibody, After COVID-19 Infection/Vaccine

COVID-19 Enfeksiyonu/Aşısından Sonra Antikora Göre Sınıflanan Kişilerde Hücresel İmmünite

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ABSTRACT

Aim: Regarding coronavirus disease (COVID-19), despite antibody measurement alone providing insufficient data, studies evaluating immune responses to vaccination or disease have focused on humoral immunity. Uncoupled humoral and cellular responses may be present after vaccination or disease. In this study, the COVID-19 recovered individuals (CRI) and vaccinated healthcare workers (VHCW) were classified into two groups according to immunoglobulin G (IgG) levels to SARS-CoV-2 spike antigen, then cellular immunity was evaluated with interferon-gamma (IFN-γ).

Material and Methods: The CRI group (n=30) had COVID-19 and were not vaccinated. The VHCW group (n=47) had two doses of CoronaVac and was never infected. In VHCW, humoral response and IFN-γ were evaluated one month after vaccination, while blood samples were taken in recovered patients between one month and one year after infection.

Results: In the VHCW group, IFN-γ (p=0.848), and age (p=0.949) were similar in IgG<7 and IgG≥7 subgroups. No correlation was present between IFN-γ and IgG levels in VHCW (p=0.711). In the CRI group, IFN-γ and age were higher in the subgroup of IgG≥7 (p=0.005, p<0.001, respectively). There was no statistically significant correlation between IFN-γ and IgG in the CRI group; however, there was a trend (p=0.057, r=0.35). No difference was observed in terms of IgG levels between the VHCW and CRI groups; while IFN-γ was higher in the CRI group (p<0.001).

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Conclusion: Demonstrating the immune response to COVID-19 is important for the development of vaccines and therapeutics. Evaluating cellular response (T cell response) to vaccines is worthy when making public health decisions during pandemics.

Key Words: SARS-CoV-2, COVID-19, Humoral Immunity, Cellular Immunity, Vaccine Immunogenicity

ÖZET

Amaç: Koronavirus hastalığında (COVID-19), antikor ölçümü tek başına yetersiz veri sağlamasına rağmen, aşılama veya hastalığa karşı immün yanıtı değerlendiren çalışmalar hümoral immüniteye odaklanmıştır. Aşılama veya hastalıktan sonra humoral ve hücresel tepkiler benzer olmayabilir. Bu çalışmada, COVID-19 geçiren bireyler (CRI) ve aşılanmış sağlık çalışanları (VHCW), SARS-CoV-2 spike antijenine karşı immünoglobulin G (IgG) seviyelerine göre iki gruba ayrıldı, ardından interferon- gama (IFN-γ) ile hücresel bağışıklık değerlendirildi.

Gereç ve Yöntem: CRI grubu (n=30) COVID-19 geçiren ve aşılanmamış bireylerden oluşmaktaydı. VHCW grubu (n=47) ise iki doz CoronaVac aşısı uygulanmış ve enfeksiyon geçirmemiş kişiler içermekteydi. VHCW grubunda, aşılamadan bir ay sonra humoral yanıt ve IFN-γ değerlendirilirken, COVID-19 geçiren hastalardan enfeksiyondan sonraki bir ay ile bir yıl arasında kan örnekleri alındı.

Bulgular: VHCW grubunda, IgG<7 ve IgG≥7 alt gruplarında IFN-γ (p=0.848) ve yaş (p=0.949) benzerdi. VHCW grubunda IFN-γ ve IgG düzeyleri arasında korelasyon saptanmadı (p=0.711). CRI grubunda, IFN-γ ve yaş, IgG≥7 alt grubunda daha yüksekti (sırasıyla p=0.005, p<0.001). CRI grubunda IFN-γ ve IgG arasında istatistiksel olarak anlamlı bir korelasyon yoktu; bununla birlikte bir trend görüldü (p=0.057, r=0.35). VHCW ve CRI grupları arasında IgG düzeyleri açısından fark saptanmadı; IFN-γ ise CRI grubunda daha yüksekti (p<0.001).

Sonuç: COVID-19'a karşı immün yanıtın gösterilmesi, aşıların ve terapötik ajanların geliştirilmesi için önemlidir. Pandemilerde halk sağlığını ilgilendiren kararlar alınırken aşı uygulamaları sonrasındaki hücresel yanıtı (T hücresi yanıtı) değerlendirmek değerlidir.

Anahtar Kelimeler: SARS-CoV-2, COVID-19, Humoral İmmünite, Hücresel İmmünite, Aşı İmmünojenitesi

INTRODUCTION

Following the emergence of the coronavirus disease 2019 (COVID-19) pandemic caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), vaccine development and production have been achieved at an unprecedented pace. In line with the efforts to achieve herd immunity after vaccinations, restrictions were eased, and vaccine responses were examined.

The first studies evaluating vaccine and disease responses mostly investigated humoral immunity, such as antibodies to spike glycoprotein or neutralizing antibodies (NAbs). However, recent studies revealed that the evaluation of humoral response alone should be questioned (1). It has been shown that antibody levels wane over time after disease or vaccination (2, 3). The new variants had a decreased susceptibility to NAbs. For instance, after the omicron variant, a new surge of infection has emerged even in countries with high vaccination rates. However, despite the reduced capacity of NAbs to variants, the clinical effects of vaccines were little affected, a finding suggesting that long-term cellular immune memory is protective against variants (1, 4). Such phenomena show that the immunological response to COVID-19 deserves to be examined beyond antibodies.

Antibody measurement alone can provide insufficient epidemiological and individual data on virus exposure. Uncoupled humoral and cellular responses may be present in acute or convalescent patients (5, 6). T-cell responses may be present in those without a humoral response, and vice versa (7, 8). Asymptomatic or mild infection may trigger T cell immunity without detectable antibodies, or with a slight antibody response (4, 9). Also, some conditions may cause decreased

humoral response (10). After vaccination, patients receiving B cell-depleting therapy exhibited a decreased antibody response; however, compared with healthy controls, their T cell responses were alike (11). In patients who did not develop the desired antibody response because of hematological malignancies, cellular responses were found to be correlated with disease severity (12). Mounting data has shown that T-cell responses are substantial for early, broad, and long-term protection from COVID-19 (1, 3, 4).

Several vaccines against SARS-CoV-2 are granted by the World Health Organization. In Turkey, the first vaccine administered to healthcare workers (HCWs) was CoronaVac (Sinovac Life Sciences, Beijing, China) which is an inactivated vaccine. Two doses of CoronaVac were administered to HCWs 28 days apart. The most appropriate way to evaluate vaccine response is to determine clinical outcomes, and there is no recommendation from the CDC to evaluate the vaccine response with antibodies. However, since HCWs are at high risk, to monitor the humoral response after vaccinization, antibodies were measured in our hospital.

Interferon-gamma (IFN-γ) is considered as a pleiotropic cytokine, boosting both the innate and adaptive immune responses to pathogens and supporting homeostasis of immune functions (13). IFN-γ is secreted predominantly by natural killer (NK), natural killer T (NKT), CD4+ T helper type 1 (Th1), CD8+ cytotoxic T, and γδ T cells. In addition, antigen-presenting cells (APCs) and B cells also secrete IFN-γ. An increase in IFN-γ indicates a Th1 response to eradicate viral infection (14). Higher IFN-γ levels were found in COVID-19 convalescent plasma (CCP) donors compared to healthy controls (15). Thus, besides NAbs, transfusion of plasma with elevated cytokines such as IFN-γ may contribute biological effects of CCP treatments (15). Insufficient IFN-γ production impairs pathogen clearance, while unrestricted IFN-γ secretion causes pathological processes (16). Early immune responses mediated by IL-17A and IFN-γ producing cells lead to faster recovery from viral infection (17). IFN-γ is a pivotal moderator of cellular immunity.

In this study, after COVID-19 infection or vaccination, both the COVID-19-recovered individuals and vaccinated HCWs were classified into two groups according to IgG levels. Afterward, cellular immunity in each group was evaluated with IFN-γ.

MATERIALS AND METHODS

The study was carried out at Polatli Duatepe State Hospital, fully complying with the principles of the Declaration of Helsinki. The participants signed an informed consent form. For this study, approvals were obtained from the Turkish Ministry of Health Scientific Research Platform (COVID-19 Scientific Research Evaluation Commission), and the Ethics Committee of Etlik Zubeyde Hanim Gynecology Training and Research Hospital (2022/15). We obtained relevant permissions for the study from the administration of Polatli Duatepe State Hospital.

Study participants

This study evaluated immune responses in COVID-19 recovered individuals (CRI) and vaccinated HCWs (VHCW). A total of 77 participants (35 female, 42 male) attended to study. The group of CRI was comprised of those who were not vaccinated and had COVID-19 (n = 30). In the CRI group, the individuals who were vaccinated before or after COVID-19 were not included in the study. There were 24 outpatients and 6 inpatients in the CRI group. VHCW group had two doses of CoronaVac and never had COVID-19 (n = 47).

Humoral and cellular response assessment

In HCWs, humoral response and IFN-γ levels were evaluated one month after two doses of vaccine. On the other hand, blood samples were taken in recovered patients with a time criterion between one month and one year

after the disease. According to the IgG level, we separated both the patients and HCWs into two groups. For classifying individuals, the antibody level cutoff was estimated as a 7 index value. The determination of the antibody cutoff was based on the level at which antibodies correlated with NAbs (18). Previously, the cutoff: 7 Index was also mentioned in another study (19).

IFN-γ levels were estimated with Human IFN-γ assay (SEA049Hu, Lot: L210224768) via ELISA (Cloud-Clone Corp., USA). Intra-assay and inter-assay CVs of IFN-γ ELISA kit were < 12%. IgG levels were measured with Siemens ADVIA Centaur SARS-CoV-2 IgG (sCOVG) assay (Ref: 11207376, Rev. 01, 2020-10) on ADVIA Centaur XPT immunoassay system (Siemens, Munich, Germany). This assay is used for the detection of IgG antibodies to the receptor binding domain of the S1 spike antigen of SARS-CoV-2 (anti-S1-RBD IgG). It was found that sCOVG levels are strongly correlated with viral-neutralizing antibody titers (20). The results considered positive > 1 Index value (U/ml), and negative below 1. According to the manufacturer, between 0.8 - 2.0 Index, within-run (repeatability), and within-laboratory (total precision) CV of the assay were ≤ 12%, and ≤ 15%, respectively. For > 2.0 Index, within-run and within-laboratory CV of the assay were ≤ 10%, and ≤ 12%, respectively.

Between 0.8 - 2.0 Index total precision that calculated in our laboratory was 11%, and for > 2.0 Index it was 5%. Within-laboratory CVs met the manufacturer's criteria.

Statistical analysis

Data analysis was carried out with SPSS for Windows 24.0 (IBM Corp., Armonk, NY, USA). Whether the continuous variables fit the normal distribution was evaluated with Kolmogorov-Smirnov and Shapiro-Wilk tests. The chi-square independence test was used to analyze the relationship between categorical variables, and Mann Whitney-U and Student's T-tests were used to compare continuous data in between-group analyses. The correlation of continuous variables was evaluated with Spearman's rho correlation. All tests of significance were 2-tailed, and p-values <0.05 were considered statistically significant.

RESULTS

In recovered patients, the median elapsed time was 120 days from the positive qRT-PCR until the blood collection, whereas for VHCW it was 30 days after the second dose of vaccination.

When the participants in VHCW were divided into 2 groups according to their IgG levels (with cutoff: 7 Index), no difference was found between these two groups in terms of IFN-γ levels, and the two groups were similar in terms of age (Table 1). No correlation was present between IFN-γ and Ab levels in the vaccine group (p = 0.711).

When CRI was divided into 2 groups according to their IgG levels (with cutoff:7), IFN-γ and age were higher in the group of Ab ≥ 7 (Table 2). There was no statistical significance for the correlation between IFN-γ and Ab in the CRI group; however, there was a trend (p = 0.057, r = 0.35). In the CRI group, the subgroup with IgG ≥ 7 Index comprised 11 individuals, and 5 of them were hospitalized patients (inpatients).

No difference was observed in terms of sex (p = 0.864), age, and Ab (p = 0.612) between the VHCW and CRI groups; while IFN-γ was higher in the CRI group (Table 3).

IgG levels of inpatients were found to be higher than outpatients (Table 4). Considering IgG levels, there were no differences between VHCW and the other two patient groups (Table 4). Inpatients' age was higher than the remaining two groups (Table 4). Inpatients and outpatients IFN-γ levels were similar (Table 4). However, both inpatients and outpatients had higher IFN-γ levels than vaccinated HCWs (Table 4).

Table 1. IFN-γ levels in vaccinated HCWs divided into two groups according to antibody level.

	VACCINATED HCWs (n = 47)		
	IgG ≥ 7 (n: 22)	**IgG < 7** (n: 25)	**p**
IFN-γ (pg/mL)	6.3 (5.3)	6.4 (4.1)	0.848
Age (years)	43 (9)	45 (11)	0.949

Data presented as Median (IQR). IgG levels were presented as Index value. HCWs - Healthcare workers

Table 2. IFN-γ levels in recovered individuals divided into two groups according to antibody level.

	COVID-19 (n = 30)		
	IgG ≥ 7 (n: 11)	**IgG < 7** (n: 19)	**p**
IFN-γ (pg/mL)	33 (127)	9.6 (6.4)	**0.005**
Age (years)	62 ± 12	40 ± 13	**< 0.001**
Days after COVID-19	120 (90)	140 (100)	0.420

Data presented as Median (IQR) or Mean ± SD. IgG levels were presented as Index value.

Table 3. IFN-γ levels in vaccinated HCWs and COVID-19 recovered individuals.

	Vaccinated HCWs (n: 47)	**Recovered individuals** (n: 30)	**p**
IFN-γ (pg/mL)	6.4 (4.4)	13.3 (24.7)	**< 0.001**
Age (years)	42 ± 8	48 ± 16	0.071

Data presented as Median (IQR) or Mean ± SD. HCWs - Healthcare workers

Table 4. IgG and IFN-γ levels in vaccinated HCWs, non-hospitalized and hospitalized patients.

	Vaccinated HCWs (n: 47)	**Outpatients** (n: 24)	**Inpatients** (n: 6)	**p**	**Post-hoc**
IgG	6.5 (7.4)	3.5 (4.9)	18.7 (18.2)	**0.024**	**p = 0.022[c]**
IFN-γ	6.4 (4.4)	10 (11.2)	30.8(108.3)	**<0.001**	**p = 0.001[a] , p < 0.001[b]**
Age(years)	42 ± 8	44 ± 14	66 ± 13	**< 0.001**	**p < 0.001[b] , p < 0.001[c]**

Data presented as Median (IQR) or Mean ± SD. IgG levels were presented as Index value. IFN-γ levels were presented as pg/mL.
a: Vaccine - outpatient; b: Vaccine - inpatient; c: Outpatient - inpatient

DISCUSSION

Here, we demonstrated that durable IFN-γ levels are present after COVID-19 infection. In recovered patients, IFN-γ levels in months later of COVID-19 were higher than IFN-γ levels measured 30 days later in those who were vaccinated, which implies after infection a robust cellular immunity response is present even after months. T-cell responses to SARS-CoV have been detected in humans even 17 years after the infection (21). Another study showed that in recovered individuals, functional SARS-CoV-2 specific immune memory persisted after mild COVID-19, and memory B and T cells persisted for at least 3 months (22). Mild SARS-CoV-2 infections can lead to prolonged immune activation even after symptoms have resolved, a finding not seen in other mild respiratory infections (23). After four months, we also saw a sustained cellular response months after the SARS-CoV-2 infection, in agreement with the aforementioned studies.

Uncoupled T and B cell responses were found in the vaccinated HCWs group. A mild cellular response occurred in vaccinated individuals 30 days after vaccination, which did not change depending on antibody levels. In addition, there was no correlation between IgG and IFN-γ levels in vaccinated HCWs. That may be because inactivated vaccines such as CoronaVac use Alum as an adjuvant, which may polarize towards a Th2 response (24, 25). We think that the lack of correlation in vaccinated individuals is due to the vaccine producing a mild cellular response and a strong antibody response after a relatively short period of one month.

In contrast, with a median time of four months after infection, the correlation between humoral and cellular immunity was borderline in recovered individuals. A finding that indicates there is better coordination of cellular and humoral responses in the disease state compared to vaccination is that IFN-γ levels are also higher in the group with high IgG levels. We think that, after infection, robust and durable T cell responses and slowly waning antibodies in months may be the reason for that kind of correlation. A recent study showed that in convalescent individuals, neutralization antibody titers correlate with the number of virus-specific T cells (26). Another study found a partial correlation between SARS-CoV-2 T cell frequencies and SARS-CoV-2 IgG ratios in moderate cases of COVID-19 (3). It seems that, in disease, clearance of the virus needs a collaborative humoral and cellular immune response.

These findings imply that there is a mild T-cell response one month after the vaccine, yet not as robust as the long-term cellular response of the infection. The interaction between B and T cell responses is crucial for effective adaptive immunity. We found that interaction, in other words, correlation, is better in recovered patients than in vaccinated individuals. This shows that after COVID-19 infection, antibody production and IFN-γ, a cellular immune marker, increase relatively together. It also shows that the cytokine response is more active in those who had the disease, compared to those who have been vaccinated. We think immune components should be in synergy to create a multi-layered defense to SARS-CoV-2 (22). We interpreted these results as a sign that the immune responses of newly developed vaccines should converge to disease responses.

T-cell responses of inactivated vaccines are considered weak. However, in theory, an advantage of inactivated vaccines is that the immune responses would target not only the spike protein of SARS-CoV-2 but also other viral proteins. Moreover, inactivated vaccines contain more conserved epitopes that may participate in T-cell responses. Similar to our study, Fu et al. revealed that, after Sinopharm and CoronaVac vaccines, a Th2-biased and a mild Th1-type response was observed (27). Some others showed that inactivated vaccines can elicit durable T-cell responses (24, 28). In addition, inactivated vaccines that use other adjuvants or combinations with Alum elicited Th1 responses (25, 29). A novel study showed that, though antibodies declined from 3 months to 12 months after CoronaVac, IFN-γ and IL-2 secretion induced by RBD and functional SARS-CoV-2 specific CD4+ and CD8+ memory T cells were up to 12 months (28). Interestingly, they also found that the cytokine profile was in favor of Th1 response rather than Th2, and mentioned that CoronaVac predominantly induces Th1-biased cellular immunity (28). We did not do such a longitudinal follow-up in our study. We think that a more detailed analysis of the Th1 and Th2 responses seen in inactivated COVID-19 vaccines is required.

In our study, the age of inpatients was found to be higher than in outpatients, which is consistent with the literature (30). While IgG levels were higher in inpatients than in

outpatients, IFN-γ levels were similar between the two patient groups. In a recent study, IFN-γ levels were the same between severe and mild disease, similar to our study (14).

Previous studies had frequently assessed cellular immunity tests such as IGRA or ELISPOT (4, 6, 10). We measured IFN-γ levels with ELISA without any stimulation of SARS-CoV-2 antigen to the samples, which may be considered a limitation. Preexisting cellular immunity primed by endemic human coronaviruses (huCOVs) may be protective for COVID-19 (3). We could not exclude whether the IFN-γ levels we found were affected by a cross-reaction of other huCOVs causing the common cold. However, we would like to point out that this is an issue that concerns not only the patients but also the vaccinated individuals in the study. In our study, we did not have a healthy control group that was both unvaccinated and uninfected. However, in the study of Ghazavi et al., they found higher IFN-γ levels in both severe and mild patients compared to healthy controls (14).

Also, there are some strengths of our study. First, we estimated IFN-γ, which is a good indicator of cellular immunity, particularly Th1 response. Second, the Siemens sCOVG assay was found to correlate well with NAbs, so we measured a marker of humoral response that could be considered a surrogate to NAbs makes the study valuable. We think it is a good approach to evaluate the cellular response after classifying the humoral response according to a certain cutoff level. In addition, the relatively small number of studies evaluating cellular responses after CoronaVac makes our study important.

CONCLUSION

In brief, by examining the immune responses against the viruses that cause pandemics, we can reveal more effective treatments for future pandemics. Demonstrating the natural immune response to infection may be important for the development of new vaccines and therapeutics. We should use tests that measure T cell response more frequently and make them widespread.

Current vaccines appear to be slightly affected by the variants. We think that we should optimize T-cell response to vaccines. By adding other immunogens to existing vaccines, cellular immunity responses may appear stronger and broader, perhaps leading to the development of pan-betacoronavirus vaccines.

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Conflict of interest

All the authors have no conflicts of interest to disclose.

Ethics approval and informed consent

The study was carried out at Polatli Duatepe State Hospital, fully complying with the principles of the Declaration of Helsinki. The participants signed an informed consent form. For this study, approvals were obtained from the Turkish Ministry of Health Scientific Research Platform (COVID-19 Scientific Research Evaluation Commission), and Ethics Committee of Etlik Zubeyde Hanim Gynecology Training and Research Hospital (2022/15). We obtained relevant permissions for the study from the administration of Polatli Duatepe State Hospital.

REFERENCES

1. Wherry EJ, Barouch DH. T cell immunity to COVID-19 vaccines. Science 2022;377(6608):821-2. https://doi.org/10.1126/science.add2897
2. Pegu A, O'Connell SE, Schmidt SD, O'Dell S, Talana CA, Lai L, et al. Durability of mRNA-1273 vaccine–induced antibodies against SARS-CoV-2 variants. Science 2021;373(6561):1372-7. https://doi.org/10.1126/science.abj4176

3. Bonifacius A, Tischer-Zimmermann S, Dragon AC, Gussarow D, Vogel A, Krettek U, et al. COVID-19 immune signatures reveal stable antiviral T cell function despite declining humoral responses. Immunity 2021;54(2):340-54. e6. https://doi.org/10.1016/j.immuni.2021.01.008
4. Vardhana S, Baldo L, Morice WG, Wherry EJ. Understanding T cell responses to COVID-19 is essential for informing public health strategies. Sci Immunol 2022;7(71):eabo1303. https://doi.org/10.1126/sciimmunol.abo1303
5. Reynolds CJ, Swadling L, Gibbons JM, Pade C, Jensen MP, Diniz MO, et al. Discordant neutralizing antibody and T cell responses in asymptomatic and mild SARS-CoV-2 infection. Sci Immunol 2020; 5(54):eabf3698. https://doi.org/10.1126/sciimmunol. abf3698
6. Schwarzkopf S, Krawczyk A, Knop D, Klump H, Heinold A, Heinemann FM, et al. Cellular immunity in COVID-19 convalescents with PCR-confirmed infection but with undetectable SARS-CoV-2–specific IgG. Emerg Infect Dis 2021;27(1):122. https://doi.org/10.3201/2701.203772
7. Broseta JJ, Rodríguez-Espinosa D, Rodríguez N, del Mar Mosquera M, Marcos MA, Egri N, et al. Humoral and cellular responses to mRNA-1273 and BNT162b2 SARS-CoV-2 vaccines administered to hemodialysis patients. Am J Kidney Dis 2021;78(4):571-81. https://doi.org/10.1053/j.ajkd.2021.06.002
8. Schiffner J, Backhaus I, Rimmele J, Schulz S, Möhlenkamp T, Klemens JM, et al. Long-term course of humoral and cellular immune responses in outpatients after SARS-CoV-2 infection. Front Public Health 2021:1378. https://doi.org/10.1101/2021.06.24.21259218
9. Gallais F, Velay A, Nazon C, Wendling M-J, Partisani M, Sibilia J, et al. Intrafamilial exposure to SARS-CoV-2 associated with cellular immune response without seroconversion, France. Emerg Infect Dis 2021;27(1):113. https://doi.org/10.3201/eid2701.203611
10. Lippi G, Henry BM, Plebani M. Optimizing effectiveness of COVID-19 vaccination: will laboratory stewardship play a role? Clin Chem Lab Med 2021;59(12):1885-8. https://doi.org/10.1515/cclm-2021-0972
11. Apostolidis SA, Kakara M, Painter MM, Goel RR, Mathew D, Lenzi K, et al. Cellular and humoral immune responses following SARS-CoV-2 mRNA vaccination in patients with multiple sclerosis on anti-CD20 therapy. Nat Med 2021;27(11):1990-2001. https://doi.org/10.1038/s41591-021-01507-2
12. Bange EM, Han NA, Wileyto P, Kim JY, Gouma S, Robinson J, et al. CD8+ T cells contribute to survival in patients with COVID-19 and hematologic cancer. Nat Med 2021;27(7):1280-9. https://doi.org/10.1038/s41591-021-01386-7
13. Lin F-C, Young HA. The talented interferon-gamma. Adv Biosci Biotechnol 2013; 4, 6-13. https://doi.org/10.4236/abb.2013.47A3002
14. Ghazavi A, Ganji A, Keshavarzian N, Rabiemajd S, Mosayebi G. Cytokine profile and disease severity in patients with COVID-19. Cytokine 2021;137:155323. https://doi.org/10.1016/j.cyto.2020.155323
15. Bonny TS, Patel EU, Zhu X, Bloch EM, Grabowski MK, Abraham AG, et al. Cytokine and chemokine levels in coronavirus disease 2019 convalescent plasma. Open Forum Infect Dis 2020 Nov 26;8(2):ofaa574. https://doi.org/10.1093/ofid/ofaa574
16. Elliott EI, Wang A. Interferon gamma runs interference on persistent COVID-19. Med (N Y) 2021;2(10):1111-3. https://doi.org/10.1016/j.medj.2021.09.004
17. Pierce CA, Preston-Hurlburt P, Dai Y, Aschner CB, Cheshenko N, Galen B, et al. Immune responses to SARS-CoV-2 infection in hospitalized pediatric and adult patients. Sci Transl Med 2020;12(564): eabd5487. https://doi.org/10.1126/scitranslmed. abd5487
18. Mulhern JG, Fadia A, Patel R, Ficociello LH, Willetts J, Dahne-Steuber IA, et al. Humoral response to mRNA versus an adenovirus vector-based SARS-CoV-2 vaccine in dialysis patients. Clin J Am Soc Nephrol 2021;16(11):1720-2. https://doi.org/10.2215/CJN.06450521
19. Spitzer A, Angel Y, Marudi O, Zeltser D, Saiag E, Goldshmidt H, et al. Association of a third dose of BNT162b2 vaccine with incidence of SARS-CoV-2 infection among health care workers in Israel. JAMA 2022;327(4):341-9. https://doi.org/10.1001/jama.2021.23641
20. Irsara C, Egger AE, Prokop W, Nairz M, Loacker L, Sahanic S, et al. Clinical validation of the Siemens quantitative SARS-CoV-2 spike IgG assay (sCOVG) reveals improved sensitivity and a good correlation with virus neutralization titers. Clin Chem Lab Med 2021;59(8):1453-62. https://doi.org/10.1515/cclm-2021-0214
21. Le Bert N, Tan AT, Kunasegaran K, Tham CYL, Hafezi M, Chia A, et al. SARS-CoV-2-specific T cell immunity in cases of COVID-19 and SARS, and uninfected controls. Nature 2020;584(7821):457-62. https://doi.org/10.1038/s41586-020-2550-z
22. Rodda LB, Netland J, Shehata L, Pruner KB, Morawski PA, Thouvenel CD, et al. Functional SARS-CoV-2-specific immune memory persists after mild COVID-19. Cell 2021;184(1):169-83. e17. https://doi.org/10.1016/j.cell.2020.11.029
23. Kennedy AE, Cook L, Breznik JA, Cowbrough B, Wallace JG, Huynh A, et al. Lasting changes to circulating leukocytes in people with mild SARS-CoV-2 infections. Viruses 2021;13(11):2239. https://doi.org/10.3390/v13112239
24. Pavel STI, Yetiskin H, Uygut MA, Aslan AF, Aydin G, Inan O, et al. Development of an inactivated vaccine against SARS CoV-2. Vaccines (Basel) 2021;9(11): 1266. https://doi.org/10.3390/vaccines9111266
25. Heinz FX, Stiasny K. Distinguishing features of current COVID-19 vaccines: knowns and unknowns of antigen presentation and modes of action. NPJ Vaccines 2021;6(1):1-13. https://doi.org/10.1038/s41541-021-00369-6
26. Ni L, Ye F, Cheng M-L, Feng Y, Deng Y-Q, Zhao H, et al. Detection of SARS-CoV-2-specific humoral and cellular immunity in COVID-19 convalescent individuals. Immunity 2020;52(6):971-7. e3. https://doi.org/10.1016/j.immuni.2020.04.023

27. Fu Y, Chen F, Cui L, Zhao Y, Zhang H, Fu S, et al. Immunological analysis of people in Northeast China after SARS-CoV-2 inactivated vaccine injection. Vaccines (Basel) 2021;9(9):1028. https://doi.org/10.3390/vaccines9091028

28. Zhao W, Chen W, Li J, Chen M, Li Q, Lv M, et al. Status of Humoral and Cellular Immune Responses within 12 Months following CoronaVac Vaccination against COVID-19. mBio 2022;13(3):e00181-22. https://doi.org/10.1128/mbio.00181-22

29. Ganneru B, Jogdand H, Daram VK, Das D, Molugu NR, Prasad SD, et al. Th1 skewed immune response of whole virion inactivated SARS CoV 2 vaccine and its safety evaluation. iScience 2021;24(4):102298. https://doi.org/10.1016/j.isci.2021.102298

30. Garcia-Beltran WF, Lam EC, Astudillo MG, Yang D, Miller TE, Feldman J, et al. COVID-19-neutralizing antibodies predict disease severity and survival. Cell 2021;184(2):476-88. e11. https://doi.org/10.1016/j.cell.2020.12.015