VIA MEDICA

ORIGINAL RESEARCH ARTICLE

Acta Haematologica Polonica 2024
Number 3, Volume 55, pages 157–167
DOI: 10.5603/ahp.99505
ISSN 0001–5814
e-ISSN 2300–7117

Evaluation of serological and hematological parameters in donors of SARS-CoV-2 convalescent plasma in respect of periods of donation

Marta Stanek[1], Anna Leśków[2*], Małgorzata Szymczyk-Nużka[1], Anita Wojciechowska-Chorębała[1], Dorota Diakowska[2]

[1]Regional Center of Transfusion Medicine and Blood Bank, Wrocław, Poland

[2]Department of Basic Sciences, Faculty of Health Science, Wroclaw Medical University, Wrocław, Poland

Abstract

Introduction: The COVID-19 pandemic has had a major impact on the health and lives of many people worldwide. According to World Health Organization (WHO) global data, 774 million cases of COVID-19 and 7.04 million COVID-19 deaths have been reported. Many people are still struggling with the consequences of infection with SARS-CoV-2. In the first months of the COVID-19 pandemic, understanding the pathophysiology of the new SARS-CoV-2 virus and establishing effective therapies became the priority. Due to the lack of dedicated treatment against SARS-CoV-2 and the limited availability of antiviral drugs, Poland was one of many countries that decided to start convalescent plasma therapy. Many COVID-19 convalescents have decided to donate whole blood and plasma.

Material and methods: In our study, we examined how the level of anti-SARS-CoV-2 antibodies changed up to 120 days after suffering from COVID-19 and developing a primary immune response, and whether the hematological parameters of convalescent donors changed significantly during this time. The study group consisted of 394 blood donors whose serum anti-SARS-CoV-2 antibody titers were analyzed via ELISA IgG assay. Additionally, hematological parameters were determined in whole blood samples.

Results: Our research shows that in blood donors who recovered from COVID-19, and whose initial anti-SARS-CoV-2 antibody levels were high, these high levels persisted even to the 120th day after the onset of the disease. Also, the hematological parameters remain normal, enabling safe blood donation for the donor and the recipient despite the previous presence of COVID-19.

Conclusions: These results can be used to optimize the parameters qualifying donors to donate convalescent plasma in the event of the future emergence of new infectious agents with significant infectivity and mortality.

Keywords: convalescent plasma, COVID-19, SARS-CoV-2, treatment, blood donor

Acta Haematologica Polonica 2024; 55, 3: 157–167

*Address for correspondence: Anna Leśków, Department of Basic Sciences, Faculty of Health Science, Wroclaw Medical University, ul. Chałubińskiego 3, 50–368 Wrocław, Poland; e-mail: anna.leskow@umw.edu.pl

Received: 22.02.2024 Accepted: 21.05.2024 Early publication date: 30.06.2024

Introduction

From the end of 2019, the world was overtaken by a pandemic caused by a new virus, severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). Starting in Wuhan, China, the pandemic quickly spread around the world, leading to the deaths of millions of people [1]. Research and clinical trials were initiated worldwide to identify effective therapeutic procedures and limit the spread of infection. Due to the need for immediate action and the fact that a targeted drug to fight the disease was unavailable, passive immunization made it possible to reduce the number of patients and the severity of the disease [2]. The European Commission for Disease Prevention and Control (ECDC) published the 'EU program of COVID-19 convalescent plasma collection and transfusion'. Treatment of patients with convalescent plasma was initiated to increase patients' chances of survival [3, 4]. Recommendations for the treatment of COVID-19 indicated the use of antiviral drugs such as remdesivir, nirmatrelvir-rytonavir, and molnupiravir, or immunomodulatory therapies such as dexamethasone and interleukin-6 or Janus kinase inhibitors [5, 6]. To reduce the spread of the virus, efforts were implemented to create an effective vaccine. Today, we know that a vaccine with mRNA or vector-based technology allows the human body to produce antibodies directed against the virus's S-protein, which prevents its entry into host cells, thereby significantly reducing the risk of developing the full disease and of spreading through the environment [7, 8]. We also know that the SARS-CoV-2 virus is constantly mutating, creating new variants, and that vaccination does not provide lifelong immunity, and therefore treatment strategies must be constantly modified [9].

Although the results of many studies indicate no reduction in mortality, and improvement but also deterioration in the health of patients after the use of COVID-19 convalescent plasma (CCP) [10, 11], convalescent plasma remains the option of treatment, especially for patients in the initial phase with severe coronavirus disease or immunocompromised patients [2, 12–14]. Moreover, convalescent plasma contains neutralizing antibodies and various anti-inflammatory molecules such as anti-inflammatory cytokines, clotting factors, natural antibodies, defensins, pentraxins and other undefined proteins [12, 15]. It must be emphasized that recovering patients are the only source of convalescent plasma [16]. In this context, convalescent plasma from donors who have been cured of infection generates a very high titer and may be particularly useful as replacement therapy in immunocompromised individuals [13, 17].

The optimal treatment strategy for CCP requires, first of all, the identification of a convalescent donor with a high titer of neutralizing antibodies [9, 14].

The aim of this study was to determine the level of anti-SARS-CoV-2 antibodies in repeat blood donors in the long term i.e. up to 120 days after confirmed virus infection. Our study aimed to evaluate hematological parameters and determine whether SARS-CoV-2 infection significantly alters hematological parameters and antibody levels depending on the time elapsed since virus infection or on demographic factors.

Material and methods

Characteristics of donors

The study group consisted of 394 blood donors who were qualified at least twice to donate convalescent plasma in the period between 27 April 2020 and 30 March 2021 in the Regional Center of Blood Donation and Blood Treatment in Wroclaw, Poland (RCBDBT-W). All data of donors was accessed from records of this institution.

The specific criteria for acceptance of the blood donors as COVID-19 convalescent plasma (CCP) donors (criteria were based on European recommendations, including European Commission Directorate — General For Health And Food Safety of 4 April 2020, developed by the National Consultant for Transfusiology's team and agreed by the public blood service continuity management working group), were as follows:

1) Donors considered healthy after recovering from COVID-19 or infection with SARS-CoV-2. Infection with SARS-CoV-2 was documented by a positive RT-PCR test result or other test result confirming infection.
2) Donors with a period of at least 14 days from two negative RT-PCR results from a nasal or pharyngeal swab, or interval of at least 28 days after the symptoms have subsided, or 18 days from the end of home isolation prior to first collection of CCP.
3) Donors with detectable antibodies against SARS-CoV-2, who were qualified to continue CCP donations.
4) Negative results of screening tests for antibodies against HLA class I, class II and HNA-antigens, which were performed in donors with a history of pregnancy or transfusion.
5) Donors who had given informed consent to participate in clinical trials.
6) Standard qualification criteria for donors of blood or its components in accordance with the Polish regulations of the Ministry of Health [18]. The main criteria were:
   a) Age: 18–65
   b) Weight: ≥50 kg
   c) Blood pressure: systolic: <180 mm Hg, diastolic: <100 mm Hg
   d) Heart rate: rhythm, with a rate of 50 to 100 beats per minute; lower heart rates are acceptable in athletic individuals with good exercise tolerance

e) Hemoglobin concentration: women: ≥125 g/L, men: ≥135 g/L
f) Without serious active, chronic or recurrent disease, and without fever.

Anti-SARS-COV-2 antibody titers

Blood samples for antibody determination were collected in special SST II Advance serum tubes with separating gel (Beckton-Dickinson, Plymouth, United Kingdom). In serum samples from each donor of CCP was performed a screening assay for detecting IgG antibodies against antigen (protein S) of the SARS-CoV-2. Anti-SARS-CoV-2 ELISA IgG assay (Euroimmun, Lubeck, Germany) was used to detect specific antibodies against the Receptor-Binding--Domain unit of SARS-CoV-2.

Euroimmun's anti-SARS-CoV-2 is an enzyme-linked immunosorbent assay (ELISA) test. The reagent wells of the ELISA were coated with an S1 domain of the spike protein of SARS-CoV-2, which reacts with specific antibodies. The assay was performed automatically using a Euroimmun Analyzer I-2P. According to the manufacturer's protocol, the absorption measurement was performed at 450 nm with a reference wavelength of 620 nm and 650 nm. Semi-quantitative evaluation of the test was made based on the manufacturer's instructions, means the ratio factor was calculated. A ratio <0.8 was considered to be non-reactive or negative, while a ratio of ≥1.1 was considered to be positive for all samples. In accordance with Polish recommendations developed by the National Consultant for Transfusiology (published on 30 June 2020), CCP was divided, according to the antibody titer, into high >500 or low <500. Plasma with a high antibody titer was defined as a multiple of the cut-off value of enzyme-linked immunosorbent tests. For the Anti-SARS--CoV-2 ELISA (IgG) Euroimmun test, this is 4.4. We assessed antibody levels at four separate periods (later named 'periods of donation'): i.e. period 1 — antibody measurement in material collected up to 60 days after donor SARS-CoV-2 infection; period 2 — material collected 61–90 days after SARS-CoV-2 infection; period 3 — material collected 91–120 days after SARS-CoV-2 infection; and period 4 — material collected more than 120 days after SARS-CoV-2 infection.

Hematological parameters assays

Hematological examination of whole blood collected to K_2-EDTA hematology tubes (Beckton–Dickinson) from each donor was performed by the use of hematology analyzers ABX Pentra XL 80 (Horiba Europe GmbH, Olomouc, Czech Republic). The following parameters were measured: quantification of red blood cells (RBCs 10^{12}/L), concentration of hemoglobin (HGB g/dL), hematocrit (Ht %), red cell distribution width (RDW %), mean corpuscular volume (MCV fL), mean corpuscular hemoglobin (MCH pg), mean corpuscular hemoglobin concentration (MCHC g/dL), quantification of white blood cells (WBCs 10^9/L), and quantification of platelets (PLT 10^9/L). Hematological parameters were also evaluated at four periods, analogous to those in which antibody levels were evaluated.

Statistical analysis

Descriptive data was presented as number of observations (percentage), mean, ± standard deviation (± SD) and 95% confidence interval (95% CI).

Data distribution was tested with a Kolmogorov–Smirnov test, equality of variances by a Levene's test, and sphericity in repeated measurements for ANOVA by a Mauchly's test.

McNemar's test was performed for analysis of categorical dependent samples. Continuous dependent data was analyzed using two-way ANOVA analysis, one-way ANOVA analysis, and then a post-hoc Tukey test. Student-t test was performed for analysis of two independent samples. Pearson correlation coefficients (r) were calculated to evaluate associations between pairs of variables. Based on Classification and Regression Tree (CART) analysis, the best-split point of age variable was identified. CART is a method of identifying predictor variables by using binary partitioning. The possible cutoff point of each variable is assessed to identify the cutoff point that results in the maximum discrimination between subgroups of patients with respect to the probability of an assessed outcome. The results of the CART analysis are presented as a 'classification tree'.

P-values of less than 0.05 were assumed to be statistically significant. Statistical analysis was performed using Statistica 13.3 software (Tibco Software Inc., Palo Alto, CA, USA).

Ethical approval

This study was performed in accordance with the Declaration of Helsinki and approved by the Bioethics Committee of Wroclaw Medical University (permission no. 536/2022).

Results

Characteristics of study group

The characteristics of the study group of convalescent plasma donors are set out in Table I. There were 353 men and 41 women with a mean age of 39.8 ± 10.6 years. The average age of women was lower than the average age of men ($p = 0.002$).

Titers of anti-SARS-CoV-2 antibodies

The total number of donations was 868 (Table I). Of these, 24.7% of blood samples showed low antibody titers, and 75.3% showed high antibody titers of anti-SARS-CoV-2 antibodies (Table II). Parameters such as gender and period of donation had no effect on the rates of high or low antibody

Table I. Characteristic of study group (n = 394)

Variable		N (%)	Mean ± SD
Sex	Men	353 (89.59)	
	Women	41 (10.41)	
Age (years)	Total		39.75 ± 10.58
	Men		40.32 ± 10.33*
	Women		34.85 ± 11.54*
Blood group	0	122 (30.96)	
	A	151 (38.32)	
	B	81 (20.56)	
	AB	40 (10.15)	
RhD factor	RhD (+)	319 (80.96)	
	RhD (–)	75 (19.04)	
Transfusion/ /pregnancy/ /multiple births	No	394 (99.74)	
	Yes	1 (0.25)	
Donations per blood donor	2	216 (54.82)	
	3	81 (20.56)	
	4	57 (14.46)	
	5	19 (4.82)	
	6 and more	21 (5.33)	
Total number of donations		868 (100.00)	

* age of men vs. age of women $p = 0.002$

titer results of anti-SARS-CoV-2 titers. Older blood donors had significantly higher titers of anti-SARS-CoV-2 antibodies than did younger donors ($p < 0.002$) (Table II).

The correlation between levels of anti-SARS-CoV-2 antibodies and the period of donation in each of the study cases was statistically significant, as set out in Fig. 1. The coefficient of correlation was 0.170 ($p < 0.0001$).

We observed insignificant differences in levels of anti-SARS-CoV-2 antibodies between the men and women subgroups (1,031.77 ± 584.52 vs. 967.41 ± 535.61; $p = 0.321$).

Because the age parameter could be a probable second factor influencing the anti-SARS-CoV-2 antibody titer, the variable dichotomization procedure was performed using CART analysis. This confirmed the age of 38 as the best cut-off point for binary partitioning of anti-SARS-CoV-2 antibody variables (Fig. 3).

Titers of anti-SARS-CoV-2 antibodies were significantly higher in the subgroup of age above 38 than in the subgroup of age below 38 (respectively 1,091.02 ± 587.32 vs. 944.08 ± 560.44, $p < 0.001$). Therefore, a two-way ANOVA analysis of levels of anti-SARS-CoV-2 antibodies for dependent variables in relation to periods of donation and age of donors was performed (Table III).

There was no effect of the age variable or of the related period of donation and age variable on the anti-antibody titer ($p > 0.05$ for both) (Table III). A significant relationship between titers of anti-SARS-CoV-2 antibodies and donation period was observed ($p = 0.008$).

As shown in Figure 4, the highest levels of anti-antibodies were observed in the 91–120 days and the above 120-day periods of donation. We demonstrated significantly lower levels of anti-SARS-CoV-2 antibodies in the up to 60 days and the 61–90 days periods of donation compared to the 91–120 days period of donation. Levels of anti-SARS-CoV-2 decreased from a 121-day period of donation, but these values were not statistically significant.

We analyzed the differences between the rates of observations of hematological parameters remaining within the norm range and those that were above or below the standard values, in selected periods of donation since a diagnosis of SARS-CoV-2 infection (Table IV). The frequency of any of the analyzed parameters did not significantly differ between selected donation periods ($p > 0.05$).

We also tested relationships between hematological parameters and periods of donation since diagnosis of SARS-CoV-2 infection (Table V). Levels of RDW and MCHC were significantly higher in Period 1 and Period 2 than in Period 3 or Period 4 ($p < 0.05$ for all), but these values were within normal ranges.

Discussion

Convalescent plasma has a long history of being used in treating infectious diseases, particularly in the absence of effective therapy and before the introduction of immunization. Convalescent plasma was used during the Spanish flu pandemic in the early 20th century and in the treatment of smallpox, measles, and hemorrhagic fever caused by Ebola virus infection [19, 20].

During the COVID-19 pandemic, due to the limited availability of COVID-19 treatment, many countries started passive immunotherapy through the use of CCP transfusion [21, 22].

Although today, more than four years after the pandemic began, the available data indicates that the use of CCP does not significantly reduce mortality or reduce the severity of symptoms in all patients with COVID-19, it is worth noting that in people with immunodeficiency and those with a severe condition in whom the current treatment is not sufficient, the use of CCP is still justified [22].

In our study, due to the fact that RCBDBT-W does not collect such data in our country, the severity of convalescent COVID-19 disease was not taken into account. However, we assumed that the symptoms of the disease were absent or mild due to (a) the quick recovery of the donors and (b) the lack of deviations from the norm in their blood parameters, which allowed them to quickly

Table II. Status of anti-SARS-CoV-2 antibodies in relation to characteristics of blood donors and number of donations Data presented as number of observations (percentage) or mean ± SD

Variables	Titer of anti-SARS-CoV-2 antibodies (n = 868)		p-value
	Low (≤500) (n = 214)	High (>500) (n = 654)	
Gender:			0.593
Men (n = 779)	190 (24.39)	589 (75.61)	
Women (n = 89)	24 (26.97)	65 (73.03)	
Age (years)	38.05 ± 9.44	40.61 ± 10.67	0.002*
Period of donation:			
Period 1 — ≤60 days	58 (26.85)	158 (73.15)	0.237
Period 2 — 61–90 days	81 (26.73)	222 (73.27)	
Period 3 — 91–120 days	30 (18.87)	129 (81.13)	
Period 4 — >120 days	45 (23.68)	145 (76.32)	

* statistically significant

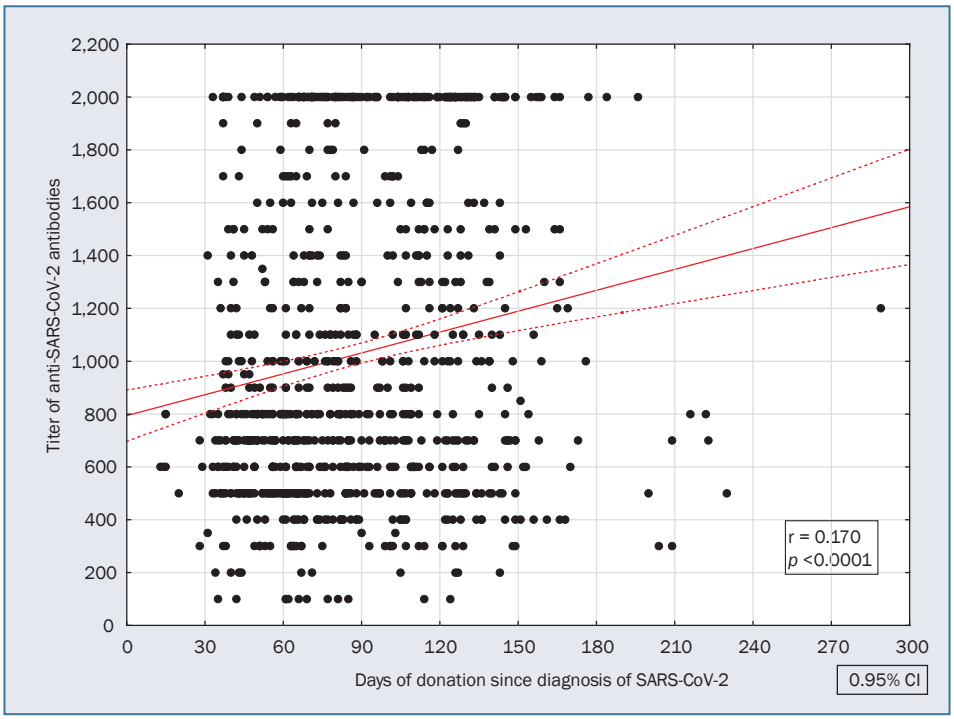

Figure 1. Correlation between titers of anti-SARS-CoV-2 antibodies and period of donation since diagnosis of disease in each study case (n = 868)

return to donation after infection with COVID-19. In accordance with the principle of donor qualification, there were no symptoms of COVID-19 on the day of qualification for blood donation. We only studied people who developed antibodies after SARS-CoV-2 infection, but not because of the vaccine. As in the studies by Skorek et al. [23] and Flisiak et al. [24], we defined high titers of specific anti-SARS-CoV-2 antibodies when titers were at least >500. A review of scientific reports about the use of convalescent plasma in the treatment of COVID-19 prepared by the Polish Agency for Health Technology Assessment and Tarification indicates that there are large differences between the titers of anti-SARS-CoV-2 antibodies in convalescent plasma used in clinical trials [25]. For example, Balcells et al. [26] found a level of antibody titers >400 to be high, yet on the other hand Salazar et al. [27] defined a high level of antibody titers as >1,350. This difference in values results from a lack of global guidelines, and the levels determined are based on the knowledge and experience of the researchers.

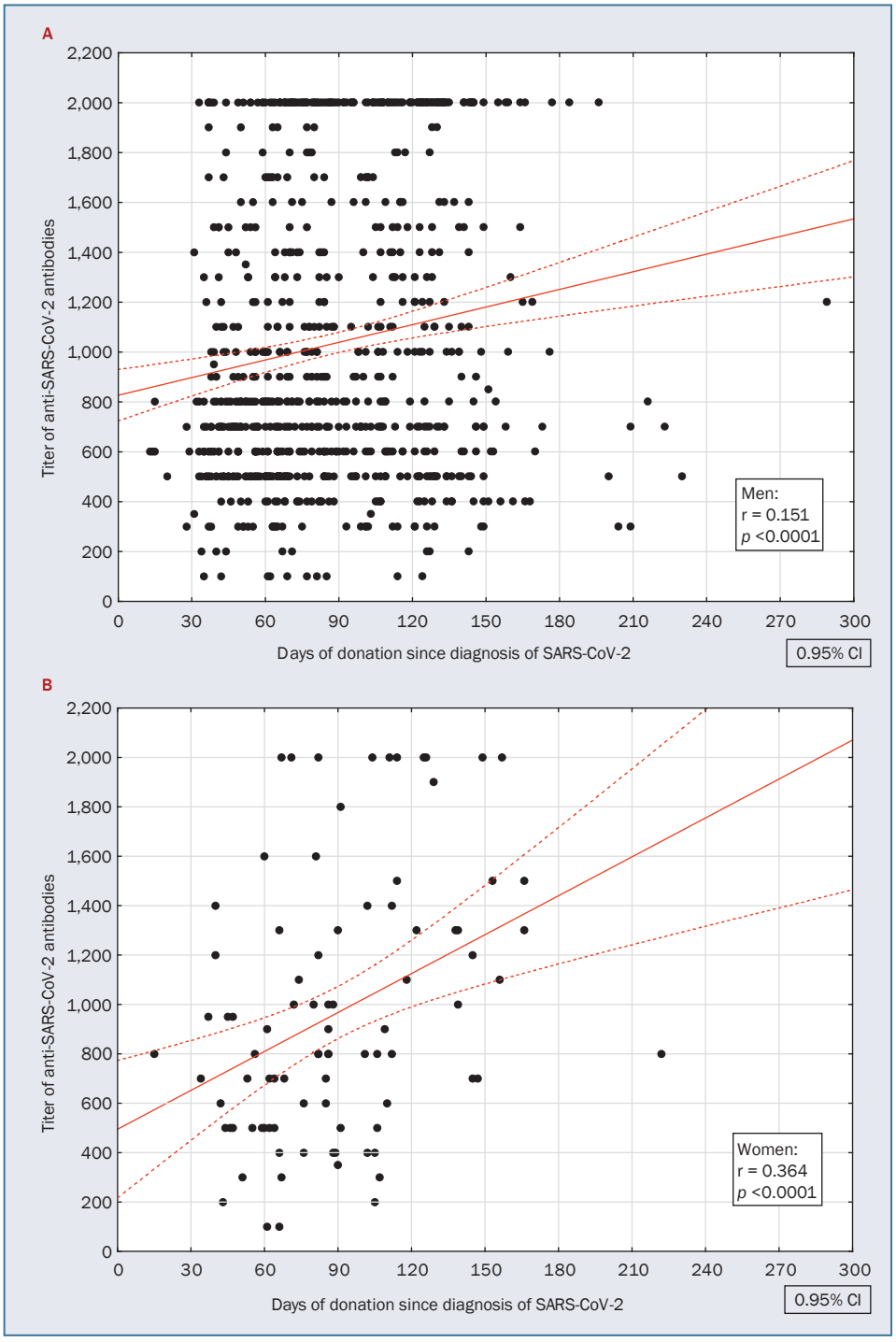

Figure 2. Correlation between titers of anti-SARS-CoV-2 antibodies and period of donation since diagnosis of disease in each case of (**A**) men subgroup (n = 779), (**B**) women subgroup (n = 89)

Based on recent reports and summaries of clinical trials [22, 25, 28], it can be concluded that standardization of guidelines regarding the titer of anti-SARS-CoV-2 antibodies that may be of clinical significance in convalescent plasma donors, especially in the first qualification test, could significantly improve the efficiency and effectiveness of COVID-19 treatment using convalescent plasma.

Our research shows that after the primary humoral immune response, the antibody titers in c.73% of examined convalescents are high for the first 90 days after the confirmation of infection, then increase in 81% of convalescents, and remain high up to 120 days. After 120 days post-SARS-CoV-2 infection confirmation, a decrease in antibodies was observed, although this tendency was not

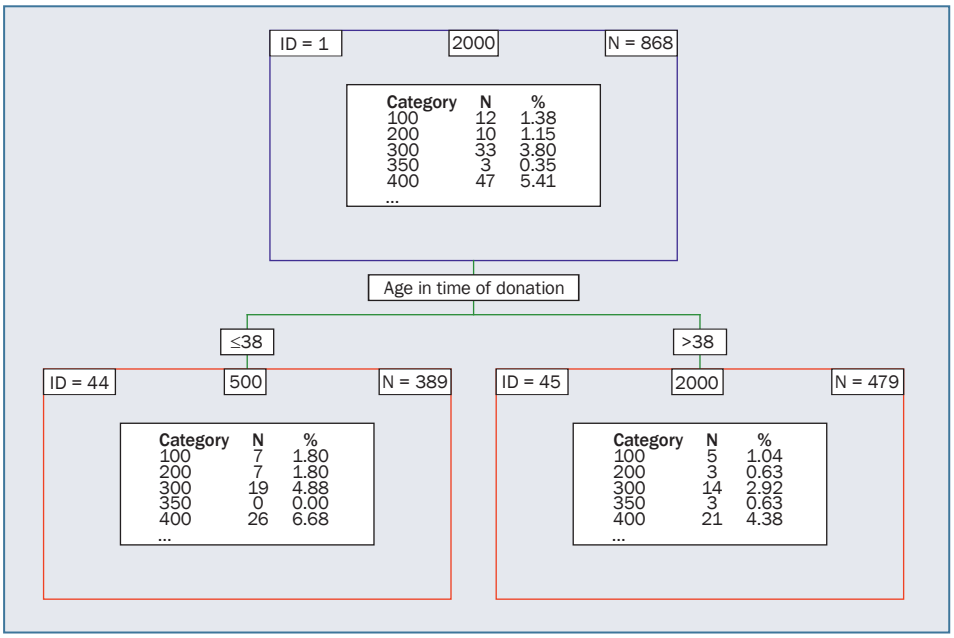

Figure 3. Classification tree for dichotomization of age variable in relation to levels of anti-SARS-CoV-2 antibody variable

Table III. Relationship between levels of anti-SARS-CoV-2 antibodies in respect of periods of donation and age of donors

Age	Period 1 ≤60 days	Period 2 61–90 days	Period 3 91–120 days	Period 4 >120 days	P-value (two-way ANOVA)		
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Period of donation	Age	Period of donation × Age
≤38 years	794.55 ± 433.87	945.45 ± 573.56	1,036.62 ± 576.74	1,046.66 ± 637.84	0.008	0.068	0.230
>38 years	903.84 ± 485.97	1,074.84 ± 601.30	1,164.21 ± 591.31	1,243.91 ± 622.11			

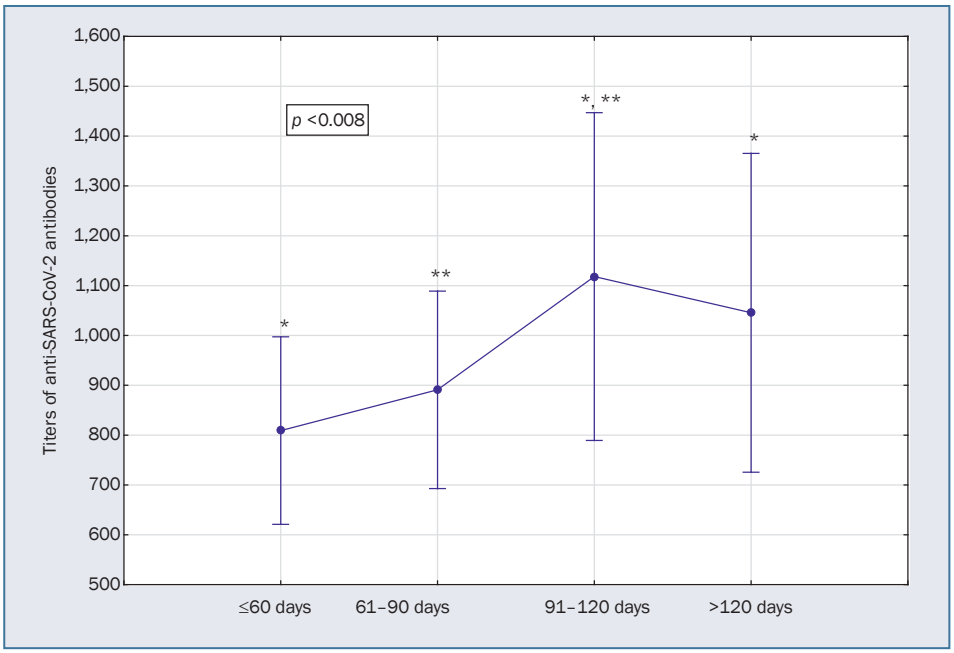

Figure 4. Levels of anti-SARS-CoV-2 antibodies in donation periods since diagnosis of SARS-CoV-2 in blood donors. Points indicate means and whiskers — confidence intervals (95% CI). * and ** $p < 0.05$

Table IV. Rates of observations of hematological parameters in relation to norm ranges, in selected periods of donation since diagnosis of SARS-CoV-2 infection. Data presented as number of observations (percentage)

Variables		Period 1 ≤60 days (n = 214)	Period 2 61–90 days (n = 303)	Period 3 91–120 days (n = 158)	Period 4 >120 days (n = 188)	P-value
RDW (%)	<11	1(0.47)	1 (0.33)	3 (1.90)	2 (1.06)	0.926
	11–16 (norm)	205 (95.79)	292 (96.37)	153 (96.83)	185 (98.40)	
	>16	8 (3.74)	10 (3.30)	2 (1.26)	1 (0.53)	
MCH (pg)	<27	3 (1.40)	7 (2.31)	4 (2.53)	3 (1.60)	0.816
	27–32 (norm)	173 (80.84)	224 (73.93)	121 (76.58)	154 (81.91)	
	>32	38 (17.76)	72 (23.76)	33 (20.89)	31 (16.49)	
MCV (fL)	<80	2 (0.93)	6 (1.98)	3 (1.90)	3 (1.60)	0.999
	80–100 (norm)	211 (98.60)	296 (97.69)	154 (97.47)	184 (97.87)	
	>100	1 (0.47)	1 (0.33)	1 (0.63)	1 (0.53)	
MCHC (g/dL)	<32	0 (0.00)	2 (0.66)	1 (0.63)	0 (0.00)	0.977
	32–36 (norm)	214 (100.00)	300 (99.00)	157 (99.37)	188 (100.00)	
	>36	0 (0.00)	1 (0.33)	0 (0.00)	0 (0.00)	
RBC (10^{12}/L)	<3.8	0 (0.00)	1 (0.33)	0 (0.00)	0 (0.00)	0.857
	3.8–6.5 (norm)	214 (100.00)	302 (99.67)	158 (100.00)	188 (100.00)	
	>6.5	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
Hematocrit (%)	<37	0 (0.00)	1 (0.33)	1 (0.63)	1 (0.53)	0.857
	37–54 (norm)	214 (100.00)	302 (99.67)	157 (99.37)	187 (99.47)	
	>54	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	
HGB (g/dL)	<11.5	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	0.772
	11.5–17.0 (norm)	213 (99.53)	292 (96.37)	154 (97.47)	182 (96.81)	
	>17.0	1 (0.47)	11 (3.63)	4 (2.53)	6 (3.19)	
WBC (10^9/L)	<4	0 (0.00)	11 (3.63)	3 (1.90)	7 (3.72)	0.594
	4–10 (norm)	206 (96.26)	288 (95.05)	151 (95.57)	179 (95.21)	
	>10	8 (3.73)	4 (1.32)	4 (2.53)	2 (1.06)	
PLT (10^9/L)	<150	1 (0.47)	6 (1.98)	2 (1.27)	3 (1.60)	0.966
	150–500	213 (99.53)	297 (98.02)	156 (98.73)	185 (98.40)	
	>500	0 (0.00)	0 (0.00)	0 (0.00)	0 (0.00)	

statistically significant. A similar tendency was reported by Ripperger et al. [29]. Higher titers of antibodies were also observed in people aged over 38, consistent with other authors' results [30–33]. On the other hand, Korper et al. [34] reported that donor factors such as gender, age, blood type (ABO), and body weight did not correlate significantly with the titer of neutralizing antibodies.

In donors participating in our study, we observed that the majority of people after full recovery from SARS-CoV-2 infection (those with a negative test for SARS-CoV-2 RNA as well as having no symptoms), obtained values of hematological parameters within the reference values. This makes such people eligible for donation, which is very important for blood collection and the use of plasma for therapeutic purposes.

Throughout the study, in all donation periods, the values of basic hematological parameters were within the reference range. This means that after the disease is sufficiently cured, the human body returns to health, and this physiologically normal condition persists. Deviations in the red blood cell system parameters were observed in a small percentage of the study group. The RDW reached higher values in the first two periods of CCP donation, which may be related to its potential predictive significance of the severity of many diseases, including viral infections. The erythrocyte survival period of 120 days allowed us to observe differences between newly formed ERCs and ERCs formed during COVID-19 infection.

Limitations of study

Our study's limitations include a focus exclusively on repeat donors, excluding those who were one-off donors or who had low initial antibody titers, and thus were not

Table V. Comparisons of hematological levels in respect of period of donation since diagnosis of SARS-CoV-2 infection. Data presented as mean + standard deviation (x + SD)

Variables	Period 1 ≤60 days	Period 2 61–90 days	Period 3 91–120 days	Period 4 >120 days	P-value (ANOVA)
RDW (%) [norm: 11–16]	13.54 ± 1.27[1, 2]	13.33 ± 1.17[3, 4]	12.82 ± 1.00[1, 3]	12.69 ± 1.01[2, 4]	<0.0001*
MCH (pg) [norm: 27–32]	30.75 ± 1.55	30.90 ± 1.65	30.84 ± 1.65	30.68 ± 1.50	0.460
MCV (fL) [norm: 80–100]	90.19 ± 4.10	90.53 ± 4.18	90.95 ± 4.05	90.44 ± 3.92	0.361
MCHC (g/dL) [norm: 32–36]	34.09 ± 0.63[1, 2]	34.12 ± 0.65[3, 4]	33.80 ± 0.63[1, 3]	33.91 ± 0.58[2, 4]	0.0001*
RBC (10^{12}/L)	4.94 ± 0.35	4.89 ± 0.37	4.92 ± 0.35	4.98 ± 0.32	0.078
Hematocrit (%) [norm: 37–54]	44.47 ± 2.64	44.27 ± 2.93[4]	44.66 ± 2.60	45.03 ± 2.82[4]	0.028*
HGB (g/dL)	15.20 ± 1.01	15.15 ± 1.12	15.14 ± 1.05	15.29 ± 1.04	0.485
WBC (10^9/L)	6.14 ± 1.32	6.13 ± 1.44	6.12 ± 1.55	6.07 ± 1.31	0.967
PLT (10^9/L)	237.33 ± 52.64	237.02 ± 49.68	237.87 ± 51.42	234.27 ± 47.59	0.903

* statistically significant; [1] Period 1 vs. Period 3, p <0.001; [2] Period 1 vs. Period 4, p <0.001; [3] Period 2 vs. Period 3, p <0.001; [4] Period 2 vs. Period 4, p <0.001

considered for subsequent CCP donations. Additionally, our analysis excluded post-vaccination antibody titer changes, focusing solely on immunity acquired from infection. Furthermore, the study did not establish detailed guidelines for the optimal timing of CCP collection or specific donor profiles, highlighting a need for future research to develop a standardized system for sample collection.

Conclusions

We conclude that natural immunity not supported by vaccination lasts up to four months, and high titers of antibodies are more likely to occur in people aged over 38.

Observations on the long-term presence of antibodies after infection with the SARS-CoV-2 virus may provide knowledge about the natural immunity of the population, as well as improve decision making on the future use of convalescent plasma treatment.

Article information and declarations

Acknowledgments

Not applicable.

Authors' contributions

MS — research concept and design, collection of data, data analysis and interpretation, writing article; AL — data analysis and interpretation, writing article; MSN and AWC — collection of data; DD — research concept and design, data analysis and interpretation, writing article. All authors — critical revision and final approval.

Conflict of interest

The authors declare no conflict of interest.

Data availability statement

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Ethics statement

All study participants have given their written consent to the use of the material collected from them for scientific purposes. Written consent was also obtained from the management of the Regional Center of Transfusion Medicine and Blood Bank to use the samples for scientific research. This study was performed in accordance with the Declaration of Helsinki and approved by the Bioethics Committee of Wroclaw Medical University (permission no. 536/2022).

Funding

The work was supported by the Regional Center of Transfusion Medicine and Blood Bank in Wrocław, Poland and by the Discipline Council of Health Sciences, Faculty of Health Sciences, Medical University in Wroclaw (grant no. REZD.Z 507.23.003).

Supplementary material

None.

References

1. Asselah T, Durantel D, Pasmant E, et al. COVID-19: Discovery, diagnostics and drug development. J Hepatol. 2021; 74(1): 168–184, doi: 10.1016/j.jhep.2020.09.031, indexed in Pubmed: 33038433.
2. Joyner MJ, Carter RE, Senefeld JW, et al. Convalescent plasma antibody levels and the risk of death from COVID-19. N Engl J Med. 2021; 384(11): 1015–1027, doi: 10.1056/NEJMoa2031893, indexed in Pubmed: 33523609.
3. Gągola K. Receiving of SARS-CoV-2/COVID-19 convalescent plasma in 2020–2021 in context of activity of Regional Center of Blood Donation and Blood Treatment in Bydgoszcz, Poland. Acta Haematol Pol. 2022; 53(5): 326–334, doi: 10.5603/ahp.a2022.0043.
4. Wang Y, Huo P, Dai R, et al. Convalescent plasma may be a possible treatment for COVID-19: A systematic review. Int Immunopharmacol. 2021; 91: 107262, doi: 10.1016/j.intimp.2020.107262, indexed in Pubmed: 33338863.
5. Andrews HS, Herman JD, Gandhi RT. Treatments for COVID-19. Annu Rev Med. 2024; 75: 145–157, doi: 10.1146/annurev-med-052422-020316, indexed in Pubmed: 37722709.
6. Yuan Y, Jiao B, Qu L, et al. The development of COVID-19 treatment. Front Immunol. 2023; 14: 1125246, doi: 10.3389/fimmu.2023.1125246, indexed in Pubmed: 36776881.
7. Park JW, Lagniton PNP, Liu Yu, et al. mRNA vaccines for COVID-19: what, why and how. Int J Biol Sci. 2021; 17(6): 1446–1460, doi: 10.7150/ijbs.59233, indexed in Pubmed: 33907508.
8. Chavda VP, Bezbaruah R, Valu D, et al. Adenoviral vector-based vaccine platform for COVID-19: current status. Vaccines (Basel). 2023; 11(2): 432, doi: 10.3390/vaccines11020432, indexed in Pubmed: 36851309.
9. Chavda VP, Bezbaruah R, Dolia S, et al. Convalescent plasma (hyperimmune immunoglobulin) for COVID-19 management: An update. Process Biochem. 2023; 127: 66–81, doi: 10.1016/j.procbio.2023.01.018, indexed in Pubmed: 36741339.
10. Iannizzi C, Chai KLi, Piechotta V, et al. Convalescent plasma for people with COVID-19: a living systematic review. Cochrane Database Syst Rev. 2023; 5(5): CD013600, doi: 10.1002/14651858.CD013600.pub6, indexed in Pubmed: 37162745.
11. Moog R. COVID 19 convalescent plasma: Is there still a place for CCP? Transfus Apher Sci. 2023; 62(2): 103680, doi: 10.1016/j.transci.2023.103680, indexed in Pubmed: 36870907.
12. Rojas M, Rodríguez Y, Monsalve DM, et al. Convalescent plasma in COVID-19: Possible mechanisms of action. Autoimmun Rev. 2020; 19(7): 102554, doi: 10.1016/j.autrev.2020.102554, indexed in Pubmed: 32380316.
13. Richier Q, De Valence B, Chopin D, et al. Convalescent plasma therapy in immunocompromised patients infected with the BA.1 or BA.2 omicron SARS-CoV-2. Influenza Other Respir Viruses. 2024; 18(3): e13272, doi: 10.1111/irv.13272, indexed in Pubmed: 38501337.
14. Filippatos C, Ntanasis-Stathopoulos I, Sekeri K, et al. Convalescent plasma therapy for COVID-19: a systematic review and meta-analysis on randomized controlled trials. Viruses. 2023; 15(3): 765, doi: 10.3390/v15030765, indexed in Pubmed: 36992474.
15. Garraud O, Heshmati F, Pozzetto B, et al. Plasma therapy against infectious pathogens, as of yesterday, today and tomorrow. Transfus Clin Biol. 2016; 23(1): 39–44, doi: 10.1016/j.tracli.2015.12.003, indexed in Pubmed: 26775794.
16. Bloch EM, Focosi D, Shoham S, et al. Guidance on the use of convalescent plasma to treat immunocompromised patients with coronavirus disease 2019. Clin Infect Dis. 2023; 76(11): 2018–2024, doi: 10.1093/cid/ciad066, indexed in Pubmed: 36740590.
17. Kumar NR, Karanam VC, Kumar S, et al. Convalescent plasma therapy in late-state, severe COVID-19 infection. South Med J. 2023; 116(5): 427–433, doi: 10.14423/SMJ.0000000000001546, indexed in Pubmed: 37137479.
18. Rozporządzenie Ministra Zdrowia w sprawie warunków pobierania krwi od kandydatów na dawców krwi i dawców krwi. Dziennik Ustaw Rzeczypospolitej Polskiej. 2017; 1741: poz. 1741.
19. Xu E, Xie Y, Al-Aly Z. Long-term neurologic outcomes of COVID-19. Nat Med. 2022; 28(11): 2406–2415, doi: 10.1038/s41591-022-02001-z, indexed in Pubmed: 36138154.
20. Pawłowska M, Styczyński J. Convalescent plasma: reaching into the past to find answers to new problems. Acta Haematol Pol. 2022; 53(5): 301–302, doi: 10.5603/ahp.2022.2040.
21. Chauhan L, Pattee J, Ford J, et al. A multicenter, prospective, observational, cohort-controlled study of clinical outcomes following coronavirus disease 2019 (COVID-19) convalescent plasma therapy in hospitalized patients with COVID-19. Clin Infect Dis. 2022; 75(1): e466–e472, doi: 10.1093/cid/ciab834, indexed in Pubmed: 34549274.
22. Casadevall A, Joyner MJ, Pirofski LA, et al. Convalescent plasma therapy in COVID-19: Unravelling the data using the principles of antibody therapy. Expert Rev Respir Med. 2023; 17(5): 381–395, doi: 10.1080/17476348.2023.2208349, indexed in Pubmed: 37129285.
23. Skorek A, Jaźwińska-Curyłło A, Romanowicz A, et al. Assessment of anti-SARS-CoV-2 antibodies level in convalescents plasma. J Med Virol. 2022; 94(3): 1130–1137, doi: 10.1002/jmv.27433, indexed in Pubmed: 34738646.
24. Flisiak R, Horban A, Jaroszewicz J, et al. Management of SARS-CoV-2 infection: recommendations of the Polish Association of Epidemiologists and Infectiologists as of April 26, 2021. Pol Arch Intern Med. 2021; 131(5): 487–496, doi: 10.20452/pamw.15979, indexed in Pubmed: 33908727.
25. Agencja Oceny Technologii Medycznych i Taryfikacji WŚOZ. Przegląd doniesień naukowych dla stosowania osocza ozdrowieńców w leczeniu COVID-19. Warszawa, 2021.
26. Balcells ME, Rojas L, Le Corre N, et al. Early versus deferred anti-SARS-CoV-2 convalescent plasma in patients admitted for COVID-19: A randomized phase II clinical trial. PLoS Med. 2021; 18(3): e1003415, doi: 10.1371/journal.pmed.1003415, indexed in Pubmed: 33657114.
27. Salazar E, Christensen PA, Graviss EA, et al. Treatment of coronavirus disease 2019 patients with convalescent plasma reveals a signal of significantly decreased mortality. Am J Pathol. 2020; 190(11): 2290–2303, doi: 10.1016/j.ajpath.2020.08.001, indexed in Pubmed: 32795424.
28. Larkey NE, Ewaisha R, Lasho MA, et al. Limited correlation between SARS-CoV-2 serologic assays for identification of high-titer COVID-19 convalescent plasma using FDA thresholds. Microbiol Spectr. 2022; 10(4): e0115422, doi: 10.1128/spectrum.01154-22, indexed in Pubmed: 35862939.
29. Ripperger TJ, Uhrlaub JL, Watanabe M, et al. Orthogonal SARS-CoV-2 serological assays enable surveillance of low-prevalence communities and reveal durable humoral immunity. Immunity. 2020; 53(5): 925–933.e4, doi: 10.1016/j.immuni.2020.10.004, indexed in Pubmed: 33129373.
30. Mehew J, Johnson R, Roberts D, et al. Convalescent plasma for COVID-19: male gender, older age and hospitalisation associated with high neutralising antibody levels, England, 22 April to 12 May

2020. Euro Surveill. 2020; 25(45): 2001754, doi: 10.2807/1560-7917.ES.2020.25.45.2001754, indexed in Pubmed: 33183404.

31. Jungbauer C, Weseslindtner L, Weidner L, et al. Characterization of 100 sequential SARS-CoV-2 convalescent plasma donations. Transfusion. 2021; 61(1): 12–16, doi: 10.1111/trf.16119, indexed in Pubmed: 32978802.
32. Gniadek TJ, Thiede JM, Matchett WE, et al. SARS-CoV-2 neutralization and serology testing of COVID-19 convalescent plasma from donors with nonsevere disease. Transfusion. 2021; 61(1): 17–23, doi: 10.1111/trf.16101, indexed in Pubmed: 32935872.
33. Klein SL, Pekosz A, Park HS, et al. Sex, age, and hospitalization drive antibody responses in a COVID-19 convalescent plasma donor population. J Clin Invest. 2020; 130(11): 6141–6150, doi: 10.1172/JCI142004, indexed in Pubmed: 32764200.
34. Körper S, Jahrsdörfer B, Corman VM, et al. Donors for SARS-CoV-2 convalescent plasma for a controlled clinical trial: donor characteristics, content and time course of SARS-CoV-2 neutralizing antibodies. Transfus Med Hemother. 2021; 48(3): 137–147, doi: 10.1159/000515610, indexed in Pubmed: 34177417.