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MAJOR ARTICLE

Persistence of Antibodies to Severe Acute Respiratory Syndrome Coronavirus 2 in Relation to Symptoms in a Nationwide Prospective Study

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(See the Editorial Commentary by Poland on pages 2163–5.)

Background. Assessing the duration of immunity following infection with severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is a first priority to gauge the degree of protection following infection. Such knowledge is lacking, especially in the general population. Here, we studied changes in immunoglobulin isotype seropositivity and immunoglobulin G (IgG) binding strength of SARS-CoV-2–specific serum antibodies up to 7 months following onset of symptoms in a nationwide sample.

Methods. Participants from a prospective representative serological study in the Netherlands were included based on IgG seroconversion to the spike S1 protein of SARS-CoV-2 (N = 353), with up to 3 consecutive serum samples per seroconverted participant (N = 738). Immunoglobulin M (IgM), immunoglobulin A (IgA), and IgG antibody concentrations to S1, and increase in IgG avidity in relation to time since onset of disease symptoms, were determined.

Results. While SARS-CoV-2–specific IgM and IgA antibodies declined rapidly after the first month after disease onset, specific IgG was still present in 92% (95% confidence interval [CI], 89%–95%) of the participants after 7 months. The estimated 2-fold decrease of IgG antibodies was 158 days (95% CI, 136–189 days). Concentrations were sustained better in persons reporting significant symptoms compared to asymptomatic persons or those with mild upper respiratory complaints only. Similarly, avidity of IgG antibodies for symptomatic persons showed a steeper increase over time compared with persons with mild or no symptoms (P = .022).

Conclusions. SARS-CoV-2–specific IgG antibodies persist and show increasing avidity over time, indicative of underlying immune maturation. These data support development of immune memory against SARS-CoV-2, providing insight into protection of the general unvaccinated part of the population.

Clinical Trials Registration. NL8473 (the Dutch trial registry).

Keywords. immunoglobulin G; COVID-19; symptoms; avidity/maturation; decay.

The persistence of specific antibodies to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), the causative agent of coronavirus disease 2019 (COVID-19), is as of yet not fully understood, partly because the follow-up time of studies investigating antibody kinetics is short owing to the novelty of the disease. Multiple studies show seroconversion to specific proteins following recent infection with SARS-CoV-2 [1–12]. Concurrently, studies report on the decay of antibodies over time, which raises the concern to what degree infected persons may remain protected to reinfection [4, 6, 8, 9, 11]. In addition, rapid decay of these antibodies would make seroprevalence estimates more difficult to interpret later after infection.

Specific antibodies are produced in different isotypes. Following most infections, immunoglobulin M (IgM) production is rapidly upregulated after infection and subsequently declines quickly [13–15]. Specific immunoglobulin A (IgA) and immunoglobulin G (IgG) antibodies typically are initiated later than IgM production. In blood, IgG is the dominant circulating antibody isotype, whereas at mucosal surfaces, including the respiratory tract, IgA antibodies are more dominant [16]. The reported decay of SARS-CoV-2 antibodies will likely differ per isotype, necessitating detailed analyses of the distribution of different antibody isotypes over longer periods of time. The presence of antibodies longer after infection, and

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rapid upregulation of antibody secretion following reinfection, depends on the presence of B-cell memory. Memory B cells are responsible for the induction of high-quality antibodies that are produced after class switching from IgM to IgG and require editing of the specificity of the antibody to provide an increased fit and binding strength of antibodies, collectively referred to as avidity maturation [17]. Hence, stronger avidity of antibodies is expected to be associated with an underlying cellular response, immune memory, and better ability to confer protection against future infection [18]. In addition to memory B cells, long-lived plasma cells contribute to the secretion of antibodies that can be detected multiple months and even years after an infection [19].

Specifically, spike S1–specific antibodies may neutralize the virus [1–3, 7, 20], for which reason many vaccines aim to induce immunity to this part of the virus [21]. Understanding of anti-spike antibody kinetics over prolonged periods of time is therefore of crucial importance [1, 5, 22, 23]. Very recent reports describe the presence of antibodies for ≥6 months after infection in specific populations such as healthcare workers or hospitalized patients [24, 25]. The duration of the antibody responses in the general population with generally mild symptoms however, has received little attention thus far.

Using samples of seroconverted individuals (N = 353) from the nationwide prospective Pienter Corona (PICO) serosurveillance study covering all ages, we studied the decay in SARS-CoV-2 spike S1–specific IgM, IgA, and IgG antibodies over a period of 7 months after infection, and investigated the effect of COVID-19–related symptoms on antibody concentrations. In addition, we studied the development of avidity of anti-SARS-CoV-2 spike S1 IgG antibodies as a marker of underlying cellular immunity and functionality of detected antibodies.

MATERIALS AND METHODS

Study Participants

Participants from the PICO serosurvey (design and inclusion are described in [7, 26, 27]) were requested to return a self-collected finger-prick blood sample in a microtainer (Sarstedt) by mail [7]. Participants were invited for a first round (PICO1) in April 2020 and for consecutive donations in June 2020 (PICO2) and October 2020 (PICO3). In the PICO2 round, the study was extended with an additional nationwide random sample [28]. Three hundred sixty-five participants seropositive for IgG to SARS-CoV-2 spike S1 were available; symptom data were missing for 12 (3.3%) participants, so 353 were included in the present study. Since we aimed to study antibodies in the general population, no other exclusion criteria were applied. Every study round, participants were asked to complete a questionnaire to collect type and date of onset of COVID-19–related symptoms data. The study was ethically approved by the Medical Research Ethics Committees United MEC-U and registered under trial number NL8473 (https://www.trialregister.nl/trial/8473). The study was performed in accordance with the Declaration of Helsinki (2008), and all participants provided written informed consent.

Laboratory Analyses

Finger-prick blood samples were centrifuged and serum stored at −20°C until analyses. The concentrations of IgG antibodies to SARS-CoV-2 spike S1 (Wuhan isolate, GenBank accession number YP_009724390.1) were determined using a fluorescent bead–based immune assay as published previously [12], which was further improved recently (Supplementary Figure 1). The assay selectively discriminates between antibodies to SARS-CoV-2 and the 4 known coronaviruses OC43, HKU-1, NL63, and 229E [12]. The specificity (99.7%) and sensitivity (91.6%) of the assay were determined using a heterogeneous sample including asymptomatic and mild to severe COVID-19 cases as representative of COVID-19 cases in the general population. Since previous publication, the assay was extended to detect IgM and IgA antibodies to spike S1 (Supplementary Figure 2). Thresholds for seropositivity were determined based on receiver operating characteristic curve analysis maximizing specificity and set at 1.20 arbitrary units (AU)/mL for IgM, 0.50 AU/mL for IgA, and 1.04 AU/mL for IgG.

Serum samples were diluted 1:200 and 1:8000 and incubated with spike S1-coupled beads in SM01 buffer (Surmodics, Eden Prairie, Minnesota) supplemented with 2% fetal calf serum while shaking (600 rpm) at room temperature for 45 minutes. Next, plates were washed 3 times in phosphate-buffered saline, incubated with phycoerythrin-conjugated anti-human IgG (Jackson ImmunoResearch Laboratories), IgA (Southern Biotech), or IgM (Jackson ImmunoResearch Laboratories) and incubated for an additional 30 minutes. Samples were washed and acquired on a LX200 or FlexMap3D (Luminex). Concentrations were interpolated from an in-house reference consisting of pooled sera using a 5-parameter logistic fit. The coefficient of variation between independent assay runs ranges from 13.3 to 17.6.

Avidity of anti-spike S1 IgG was performed on 73 samples of randomly selected participants with varying concentrations of IgG by testing samples within the linear range of detection in the absence or presence of 1.1 M of the chaotropic agent ammonium-thiocyanate [29, 30]. This concentration was confirmed to provide an optimal balance in discriminating antibodies of low and high avidity. Avidity is expressed as percentage of binding remaining when ammonium-thiocyanate is added.

Statistical Analyses

All statistical analyses were performed in R version 4.0.2 [31]. Participants with fever, dyspnea, muscle ache, extreme tiredness, general malaise, painful respiration, joint pain, diarrhea, and/or stomach ache were considered symptomatic for COVID-19. Asymptomatic participants and participants with mild upper respiratory tract complaints only (runny nose, sore throat, anosmia/ageusia, headache) were grouped together since these symptoms suggest contained, nonprogressive infection. Sera of 365 participants were available, of which 12 were excluded because symptom data were missing.

Days since onset of symptoms for symptomatic and mildly symptomatic participants was defined as the number of days between symptom onset and the blood collection date. For asymptomatic participants, the mean number of days since onset of symptoms of symptomatic persons was used as a surrogate measure to calculate their days since infection. To show seropositivity over time, time since onset of symptoms was categorized into month 1 (0–30 days)—the period of induction of antibody production—and subsequently in months 2–3 (31–92 days), months 4–5 (93–152 days), and ≥6 months (>152 days).

To study the change in the antibody concentrations and IgG avidity over time, antibody concentrations (AU/mL) were natural log-transformed and modeled separately. For each isotype, participants were included based on evidence of seroconversion to exclude persons who did not convert for IgM or IgA to influence decay rates (Supplementary Table 1). For IgG avidity, all available data were used. Generalized estimating equations with an exchangeable correlation structure was used to take into account correlation due to repeated sampling (using *geepack* version 1.3.1 [32–34]). We selected the model with exponential decay over time if it resulted in a decrease in QIC (quasi-likelihood under independence model criterion) of at least 2 compared to a model with a linear change over time [35]. Hereafter, age, sex, days since onset of symptoms, presence and duration of symptoms, and an interaction term between days since onset of symptoms and symptoms were included in the model as potential predictor variables. Age and duration of symptoms were dichotomized at their median (ie, ≥50 vs ≤49 years of age and ≥11 vs ≤10 days, respectively). Variables with $P < .100$ in univariable analyses were included in the multivariable model. Backwards selection was performed manually, excluding variables one-by-one with $P > .050$. Reported *P* values are from model coefficients. The 2-fold decrease of IgG antibodies was calculated using the slope estimate and its 95% confidence interval (CI) (ie, -log 2/slope) [29].

RESULTS

Description of the Study Population

Sera of 353 participants with specific IgG antibodies to spike S1 were available for analysis (Figure 1A). In total, 738 samples of these participants were analyzed, which are shown relative to date of onset of symptoms in Figure 1B.

The majority of participants reported a date of onset of symptoms that was close to the peak of the first wave of COVID-19 infections in the Netherlands [36]. Of the 353 participants, 214 reported symptoms and 139 reported no (n = 77) or only very mild (n = 62) upper respiratory tract symptoms (Table 1). The median age was 48 years (interquartile range [IQR], 30–61 years) and 51 years (IQR, 32–66 years) for symptomatic and asymptomatic/mildly symptomatic persons, respectively. Of the symptomatic and asymptomatic/mildly symptomatic participants, 60% and 53%, respectively, were female. The most frequently reported symptoms were headache (67%), coughing (63%), fever (57%), muscle ache (52%), and general malaise (49%), while 35% reported dyspnea. Forty percent of those from the symptomatic participant group visited the general practitioner and 2% were admitted to the hospital.

Seropositivity to IgM, IgA, and IgG Anti-Spike S1

The majority of individuals had anti-spike S1 IgM (64%) and IgA (62%) antibodies in the first month after SARS-CoV-2 IgG seroconversion (Figure 2A). The proportion of IgM- and IgA-positive participants decreased after the first month to approximately 50% at 2–3 months after onset of symptoms. After 6 months since onset of symptoms, 33% (95% CI, 28%–39%) and 37% (95% CI, 31%–43%) remained positive for IgM and IgA, respectively. In the first month, 99% of the participants were IgG positive, which increased to 100% in months 2–3. After 6 months, 92% (95% CI, 89%–95%) were still positive for IgG.

Seropositivity in Relation to Symptoms

Symptomatic individuals were more frequently positive for IgM or IgA in the first month after SARS-CoV-2 IgG seroconversion (Figure 2B and 2C; Supplementary Table 2*A*) compared with asymptomatic/mildly symptomatic persons. This difference gradually decreased over time, though it was still present after 6 months with 10% and 14% more symptomatic participants being positive for IgM and IgA, respectively, compared with asymptomatic/mildly symptomatic persons. IgG anti-spike S1 seropositivity was observed regardless of COVID-19 symptoms. However, after 6 months, the individuals who had turned negative for IgG were mostly asymptomatic/mildly symptomatic: 87% positive for asymptomatic/mildly symptomatic persons vs 95% positive for symptomatic persons (Figure 2D; Supplementary Table 2*A*).

Concentrations of Anti-Spike S1 Antibodies Over Time in Relation to Symptoms

Among persons who seroconverted to spike S1 IgM (n = 86), IgM concentrations showed a linear decline over time and initially were higher in symptomatic persons than asymptomatic/mildly symptomatic persons, but were similar from 2 months post onset of symptoms onward (Figure 3A; Supplementary Table 2*B*). The average concentration

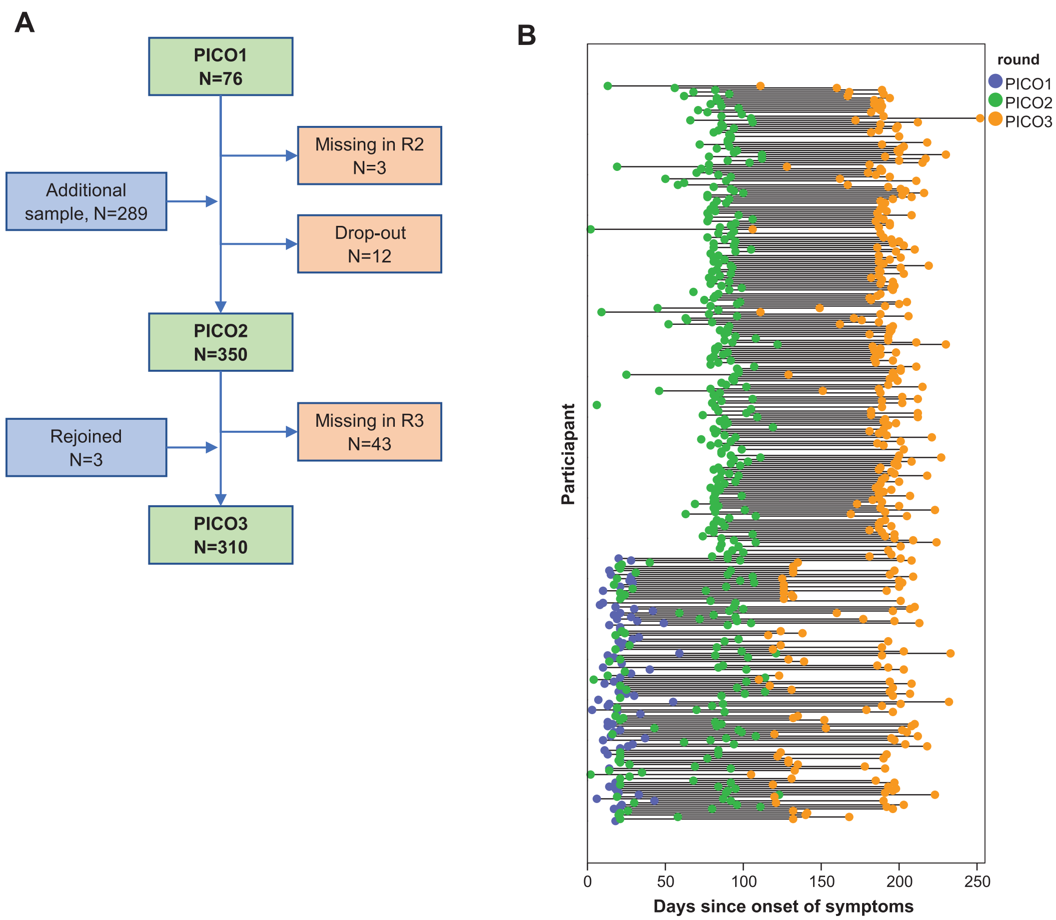

Figure 1. *A*, Flow diagram of number of participants throughout the study. *B*, The availability of consecutive samples from the 3 Pienter corona (PICO) rounds relative time since onset of disease to days since onset of symptoms (x-axis). Each line represents a participant, with the dot indicating the days since onset of disease and the lines the availability of consecutive samples.

of IgM decreased to the threshold for seropositivity after around150 days. Among persons who seroconverted to spike S1 IgA (n = 82), IgA concentrations showed an exponential decrease over time (Figure 3B). The presence of symptoms resulted in higher IgA concentrations (Supplementary Tables 2B, 2C, and 3). Average IgA concentration reached the threshold concentration after around 140 days. IgG concentrations showed a linear decrease over time, and symptomatic persons had significantly higher concentrations (Figure 3C; Supplementary Table 2*B*). The average concentrations of IgG did not intersect the threshold value for seropositivity within the studied time frame of 7 months after onset of symptoms. IgM and IgA antibody concentrations over time for the entire study population—including those who did not seroconvert to IgM and IgA in the first 60 days following symptom onset—are shown in Supplementary Table 2*C*. IgG and IgA, but not IgM, levels were higher in males and persons older than 50 years (Supplementary Table 3). In addition, duration of symptoms for longer than 10 days resulted in increased IgG levels.

Decrease in Concentration and Avidity Maturation of IgG Anti-Spike S1

Since IgG antibodies persist, we calculated the 2-fold decrease and measured avidity for IgG. The 2-fold decrease of IgG concentrations, corrected for age, sex symptoms, and duration of symptoms, was estimated to be 158 days (95% CI, 136–189 days). In addition to the duration of IgG in serum, we assessed the maturation of IgG to spike S1 by assessing the avidity. The avidity index of spike S1–specific IgG antibodies increased >2-fold during the 7 months after onset of symptoms ($P < .015$; Figure 3D). Symptomatic individuals showed a stronger increase over time than asymptomatic/mildly symptomatic individuals ($P = .022$; Supplementary Table 3).

DISCUSSION

In light of the urgent question of the duration of immunity to SARS-CoV-2 following infection in the general population, we systematically studied the dynamics in seropositivity and concentrations of IgM, IgA, and IgG antibodies to the SARS-CoV-2 spike S1 protein among cases with different symptom profiles and investigated IgG maturation over time. Our data confirm

Table 1. Characteristics of Seroconverted Individuals

Characteristic	Symptomatic	Asymptomatic/ Only Mild Symptoms
No. of participants	214	139
Symptoms		
Runny nose	48% (103)	17% (23)
Sore throat	37% (79)	11% (15)
Cough	63% (135)	19% (27)
Ageusia/anosmia	46% (98)	13% (18)
Headache	67% (144)	14% (20)
Fever	57% (133)	NA[a]
Dyspnea	35% (74)	NA[a]
Muscle ache	52% (112)	NA[a]
Extreme fatigue	34% (73)	NA[a]
Painful respiration	16% (34)	NA[a]
Diarrhea	29% (61)	NA[a]
Joint pain	24% (52)	NA[a]
Stomach ache	21% (44)	NA[a]
General malaise	49% (104)	NA[a]
No symptoms	NA	56% (77)
Age, y, median (IQR)	48 (30–61)	51 (32–66)
Male sex	40% (85)	47% (65)
Duration of symptoms,[b] median (IQR)	11 (6–18)	6 (2–9)

Data are presented as % (no.) unless otherwise indicated.

Abbreviations: IQR, interquartile range; NA, not applicable.

[a]Participants with these symptoms are included in the "symptomatic" group and therefore shown as "NA" in the "asymptomatic/only mild respiratory symptoms" group.

[b]Data on the duration of symptoms were available for 153 participants in the symptomatic group and 26 participants in the asymptomatic/only mild upper respiratory symptoms group.

that antibodies decline rapidly in the case of IgM and IgA isotypes. In contrast, 87% of the asymptomatic/mildly symptomatic and 95% of the symptomatic participants remained positive for IgG 7 months after onset of COVID-19 symptoms. Moreover, the estimated 2-fold decrease in concentration of 158 days and the increasing avidity of anti-spike IgG antibodies indicate the presence of memory B cells and/or long-lived plasma cells.

We showed that IgM and IgA antibodies start to decay within a few months after onset of symptoms, which may help explain the decline in seropositivity in some studies [6, 11, 13–15]. Since IgG antibodies persist much longer than IgM and IgA antibodies, the detection of IgG provides better sensitivity longer after infection, and therefore, IgG should be the isotype of choice in studies aiming to assess seroprevalence >2 months after the infection and in longitudinal studies. IgG may also be the most informative for identifying memory induction, since specific IgG antibody development requires multiple cell divisions and class-switch recombination, processes that are a hallmark of memory formation. The hallmarks of memory formation—IgG antibodies with high avidity and persistence of antibodies—are presented in this study. The 2-fold decrease of IgG estimated in this study was 5- to 6-fold longer than the decay of passively transferred maternal antibodies [29, 37, 38]. This decrease rate may still be underestimated since the decay

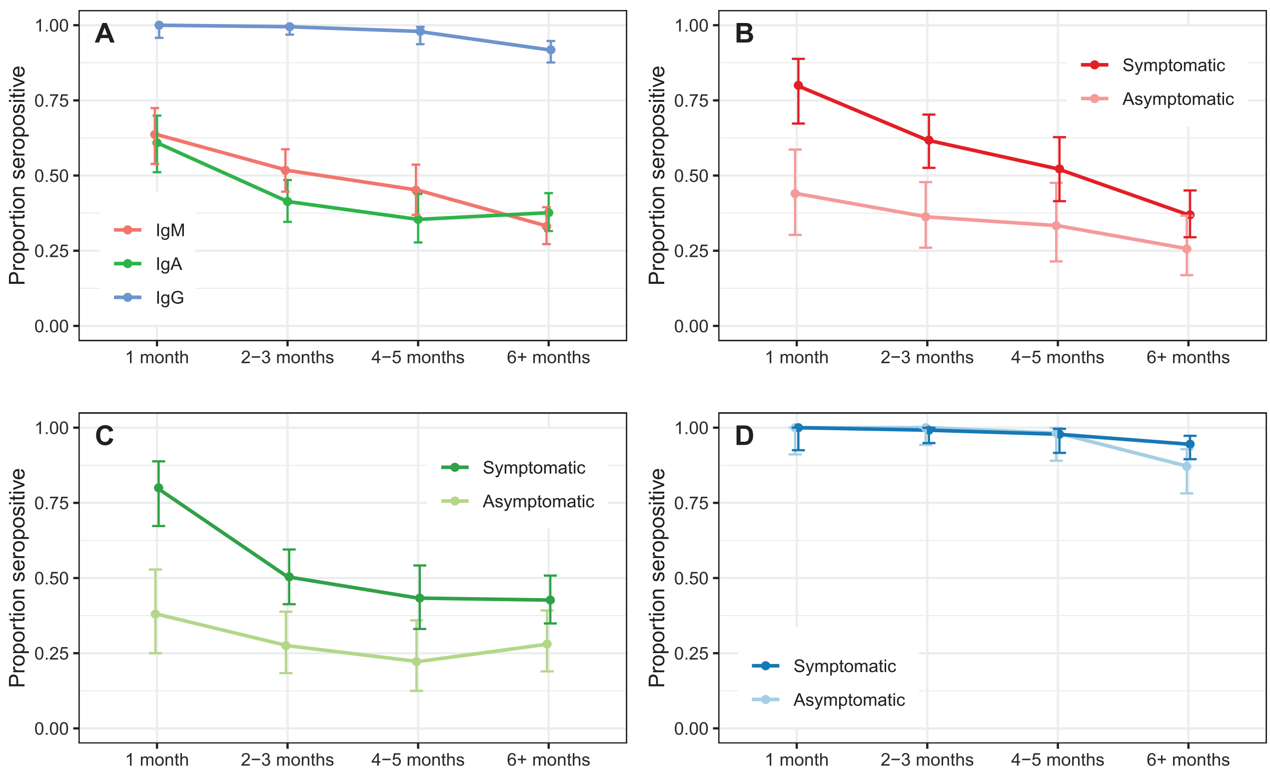

Figure 2. *A*, The proportion of immunoglobulin M (IgM), immunoglobulin A (IgA), and immunoglobulin G (IgG) and 95% confidence intervals (CIs) of positive samples in relation to months since onset of symptoms. The proportion of individuals positive for IgM (*B*), IgA (*C*), and IgG (*D*) with symptoms, or with mild or no symptoms.

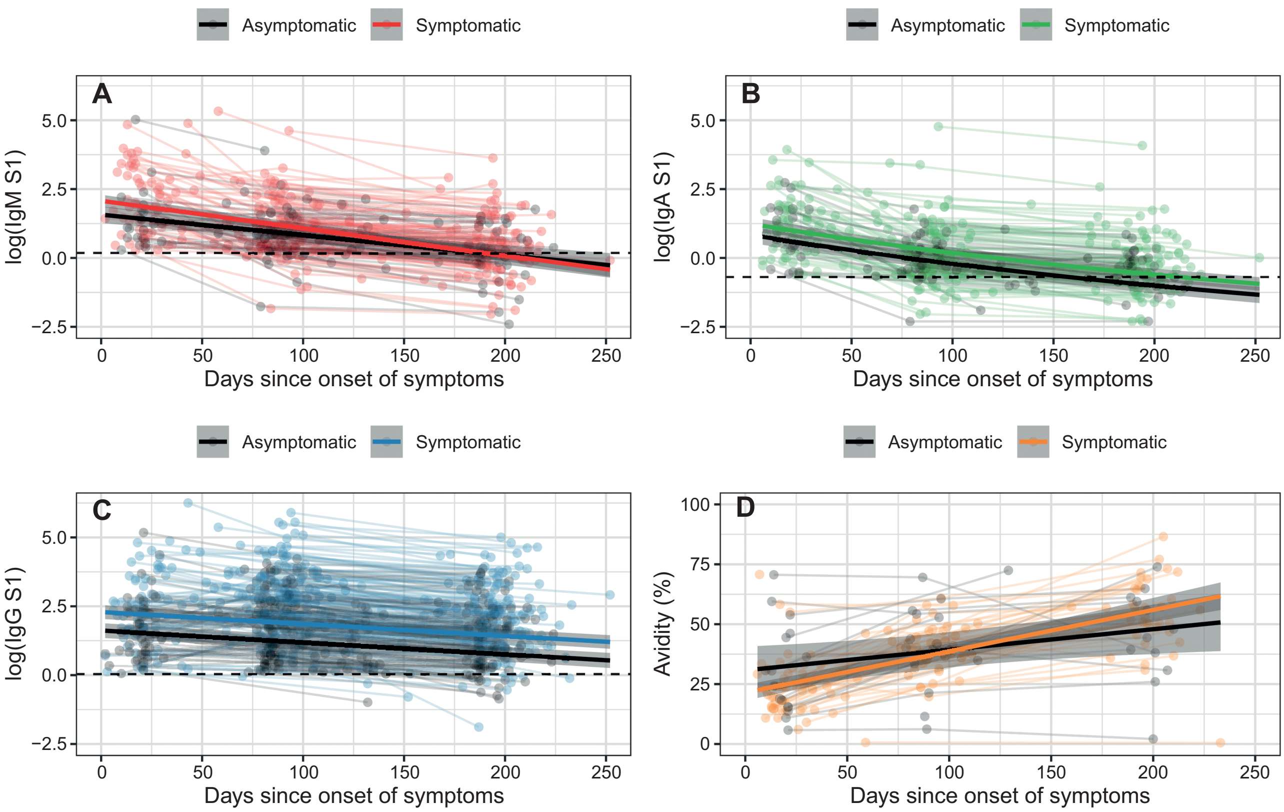

Figure 3. The concentrations of immunoglobulin M (IgM, *A*), immunoglobulin A (IgA, *B*), and immunoglobulin G (IgG, *C*) are shown relative to days since onset of symptoms for individuals having symptoms (colored lines) or those without or only mild symptoms (black lines). *D*, Development of IgG avidity for persons with or without symptoms. Data were fitted using generalized estimating equations and show 95% CIs around the fit, with an exponential decay over time for IgA and a linear relationship for IgM, IgG, and avidity. The fit was adjusted for age, sex, symptoms, and the duration of symptoms where appropriate (Supplementary Table 3). For IgM, univariable regression analysis did not show an association between symptoms and IgM levels (ie, $P > .10$; see Methods) but results by group are shown here for consistency. Transparent dots and connected lines represent (repeated) measures per individual.

of antibodies is the most pronounced in the first months after the induction of the antibodies. Therefore, longer follow-up studies should reassess the persistence of antibodies to spike S1 of SARS-CoV-2 and compare these to persistence as observed for other viruses [39, 40].

The formation of B-cell memory implies that antibodies can be rapidly upregulated in response to reinfection in order to effectively control the virus [18, 41]. It is still unknown which antibody levels confer protection against reinfection or COVID-19 disease. While the antibodies detected in this study are restricted to spike S1, we cannot exclude the detection of antibodies not necessarily contributing to virus neutralization. In light of newly emerging strains with mutations that may escape neutralization by antibodies, the cross-protection by preexisting immunity, either through infection or vaccination, needs to be closely monitored. Interestingly, having had COVID-19–like symptoms resulted in higher antibody concentrations for IgG and IgM and faster development of IgG avidity, compared with persons who remained asymptomatic/mildly symptomatic after SARS-CoV-2 infection. The reason for this may be a stronger inflammatory response, a higher or longer viral replication period, or both, that may result in better and longer-lasting immunity.

This study is unique in analyzing samples collected in the general population including all ages and COVID-19 disease severities. While the findings reflect SARS-CoV-2 antibody dynamics of the general public, the study has several limitations. Participants were included based on IgG anti-spike S1 seropositivity, and therefore we may have missed a few persons who seroconverted for IgM or IgA, but not, or insufficiently, for IgG. The time since onset of COVID-19 was based on self-reported symptoms on a presumed SARS-CoV-2 infection, and therefore may be less accurate since symptoms could be caused by other infections still prevailing during the peak of the epidemic. However, the reported date of onset of symptoms of the participants matched the national epidemiological data of COVID-19 cases in the Netherlands [36]. In addition, the paired samples of seroconverted individuals collected 6 months apart confirm that IgG antibodies persist for >6 months in 92% of seroconverted individuals [42]. Despite the persistence of IgG antibodies, the decay cannot be neglected and will eventually result in an underestimation of the proportion of infected persons in the population once this proportion has crossed the cutoff levels of specific antibody detection.

In conclusion, our analyses included 353 individuals participating in a nationwide population study with 7 months' follow-up for most participants, which is a substantially longer follow-up period than most other population studies [3, 10]. We show that anti-SARS-CoV-2 spike S1 IgG antibodies persist for an extended time (ie, >6 months). Therefore, we propose that analysis of IgG anti-spike S1 of SARS-CoV-2 will generate the most consistent seroprevalence estimates and provide understanding of the duration of protective immunity. In view of an IgG decay rate 5- to 6-fold slower than reported for passively transferred maternal IgG and the improving IgG avidity over time, B-cell memory is likely established in most individuals. In addition, our data suggest that the duration of the IgG response is likely longer for symptomatic COVID-19 cases due to higher initial concentrations. Our results aid the interpretation of the duration of immunity in unvaccinated persons and provide a framework for the evaluation of immunity induced by vaccines for SARS-CoV-2.

Supplementary Data

Supplementary materials are available at *Clinical Infectious Diseases* online. Consisting of data provided by the authors to benefit the reader, the posted materials are not copyedited and are the sole responsibility of the authors, so questions or comments should be addressed to the corresponding author.

Notes

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Potential conflicts of interest. The authors: No reported conflicts of interest. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest.

References

1. Lin Q, Zhu L, Ni Z, Meng H, You L. Duration of serum neutralizing antibodies for SARS-CoV-2: lessons from SARS-CoV infection. J Microbiol Immunol Infect **2020**; 53:821–2.
2. Crawford KHD, Dingens AS, Eguia R, et al. Dynamics of neutralizing antibody titers in the months after SARS-CoV-2 infection. J Infect Dis **2020**; doi:10.1093/infdis/jiaa618.
3. Gudbjartsson DF, Norddahl GL, Melsted P, et al. Humoral immune response to SARS-CoV-2 in Iceland. N Engl J Med **2020**; 383:1724–34.
4. Zhang G, Nie S, Zhang Z, Zhang Z. Longitudinal change of severe acute respiratory syndrome coronavirus 2 antibodies in patients with coronavirus disease 2019. J Infect Dis **2020**; 222:183–8.
5. Wang K, Long Q-X, Deng H-J, et al. Longitudinal dynamics of the neutralizing antibody response to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection. Clin Infect Dis **2021**; 72:e531–e539.
6. Seow J, Graham C, Merrick B, et al. Longitudinal observation and decline of neutralizing antibody responses in the three months following SARS-CoV-2 infection in humans. Nat Microbiol **2020**; 5:1598–607.
7. Vos ERA, den Hartog G, Schepp RM, et al. Nationwide seroprevalence of SARS-CoV-2 and identification of risk factors in the general population of the Netherlands during the first epidemic wave. J Epidemiol Commun Health **2020**; doi:10.1136/jech-2020–215678.
8. Wang X, Guo X, Xin Q, et al. Neutralizing antibody responses to severe acute respiratory syndrome coronavirus 2 in coronavirus disease 2019 inpatients and convalescent patients. Clin Infect Dis **2020**; doi:10.1093/cid/ciaa/721.
9. Iyer AS, Jones FK, Nodoushani A, et al. Persistence and decay of human antibody responses to the receptor binding domain of SARS-CoV-2 spike protein in COVID-19 patients. Sci Immunol **2020**; 5:eabe0367.
10. Isho B, Abe KT, Zuo M, et al. Persistence of serum and saliva antibody responses to SARS-CoV-2 spike antigens in COVID-19 patients. Sci Immunol **2020**; 5:eabe5511.
11. Ibarrondo FJ, Fulcher JA, Goodman-Meza D, et al. Rapid decay of anti–SARS-CoV-2 antibodies in persons with mild covid-19. N Engl J Med **2020**; 383:1085–7.
12. den Hartog G, Schepp RM, Kuijer M, et al. SARS-CoV-2-specific antibody detection for seroepidemiology: a multiplex analysis approach accounting for accurate seroprevalence. J Infect Dis **2020**; 222:1452–61.
13. Welliver RC, Kaul TN, Putnam TI, Sun M, Riddlesberger K, Ogra PL. The antibody response to primary and secondary infection with respiratory syncytial virus: kinetics of class-specific responses. J Pediatr **1980**; 96:808–13.
14. Hsueh PR, Huang LM, Chen PJ, Kao CL, Yang PC. Chronological evolution of IgM, IgA, IgG and neutralisation antibodies after infection with SARS-associated coronavirus. Clin Microbiol Infect **2004**; 10:1062–6.
15. Panyasing Y, Goodell CK, Giménez-Lirola L, et al. Kinetics of influenza A virus nucleoprotein antibody (IgM, IgA, and IgG) in serum and oral fluid specimens from pigs infected under experimental conditions. Vaccine **2013**; 31:6210–5.
16. Macpherson AJ, McCoy KD, Johansen FE, Brandtzaeg P. The immune geography of IgA induction and function. Mucosal Immunol **2008**; 1:11–22.
17. Stavnezer J, Guikema JE, Schrader CE. Mechanism and regulation of class switch recombination. Annu Rev Immunol **2008**; 26:261–92.
18. Inoue T, Moran I, Shinnakasu R, Phan TG, Kurosaki T. Generation of memory B cells and their reactivation. Immunol Rev **2018**; 283:138–49.
19. Hammarlund E, Thomas A, Amanna IJ, et al. Plasma cell survival in the absence of B cell memory. Nat Commun **2017**; 8:1781.
20. Rogers TF, Zhao F, Huang D, et al. Isolation of potent SARS-CoV-2 neutralizing antibodies and protection from disease in a small animal model. Science **2020**; 369:956–63.
21. Amanat F, Krammer F. SARS-CoV-2 vaccines: status report. Immunity **2020**; 52:583–9.
22. den Hartog G, van Binnendijk R, Buisman AM, Berbers GAM, van der Klis FRM. Immune surveillance for vaccine-preventable diseases. Expert Rev Vaccines **2020**; 19:327–39.
23. Jiang S, Hillyer C, Du L. Neutralizing antibodies against SARS-CoV-2 and other human coronaviruses. Trends Immunol **2020**; 41:355–9.
24. Lumley SF, Wei J, O'Donnell D, et al. The duration, dynamics and determinants of SARS-CoV-2 antibody responses in individual healthcare workers. Clin Infect Dis **2021**; 72:e699–e709.
25. Dan JM, Mateus J, Kato Y, et al. Immunological memory to SARS-CoV-2 assessed for up to 8 months after infection. Science **2021**:eabf4063.
26. Verberk JDM, Vos RA, Mollema L, et al. Third national biobank for population-based seroprevalence studies in the Netherlands, including the Caribbean Netherlands. BMC Infect Dis **2019**; 19:470.
27. Vos ERA, van Boven M, den Hartog G, et al. Associations between measures of social distancing and SARS-CoV-2 seropositivity: a nationwide population-based study in the Netherlands. medRxiv [Preprint]. February 12, **2021**. Available from: doi:10.1101/2021.02.10.21251477.
28. Pienter corona study team. Bilthoven, the Netherlands: National Institute for Public health and the Environment, **2020**. Available at: https://www.rivm.nl/en/pienter-corona-study.
29. Berbers G, Mollema L, van der Klis F, den Hartog G, Schepp R. Antibody responses to respiratory syncytial virus: a cross-sectional serosurveillance study in the Dutch population focusing on infants younger than 2 years. J Infect Dis **2020**; doi:10.1093/infdis/jiaa483.
30. Scherpenisse M, Schepp RM, Mollers M, Meijer CJ, Berbers GA, van der Klis FR. Characteristics of HPV-specific antibody responses induced by infection and vaccination: cross-reactivity, neutralizing activity, avidity and IgG subclasses. PLoS One **2013**; 8:e74797.
31. R Project for Statistical Computing. R: a language and environment for statistical computing. Vienna, Austria: R Foundation, **2013**.
32. Halekoh U, Højsgaard S, Yan J. The R package geepack for generalized estimating equations. J Stat Softw **2006**; 15:1–11.
33. Yan J, Fine J. Estimating equations for association structures. Stat Med **2004**; 23:859–74; discussion 875–7, 879–80.
34. Yan J. Geepack: yet another package for generalized estimating equations. R-news **2002**; 2:12–4.

35. Burnham KP, Anderson DR. Model selection and multimodel inference: a practical information-theoretic approach. 2nd ed. New York: Springer-Verlag, **2002**.
36. National Institute for Public Health and the Environment. Current information about COVID-19 (novel coronavirus). Available at: https://www.rivm.nl/en/novel-coronavirus-covid-19/current-information. Accessed 15 September 2020.
37. Sarvas H, Seppälä I, Kurikka S, Siegberg R, Mäkelä O. Half-life of the maternal IgG1 allotype in infants. J Clin Immunol **1993**; 13:145–51.
38. Waaijenborg S, Hahné SJ, Mollema L, et al. Waning of maternal antibodies against measles, mumps, rubella, and varicella in communities with contrasting vaccination coverage. J Infect Dis **2013**; 208:10–6.
39. Amanna IJ, Carlson NE, Slifka MK. Duration of humoral immunity to common viral and vaccine antigens. N Engl J Med **2007**; 357:1903–15.
40. Antia A, Ahmed H, Handel A, et al. Heterogeneity and longevity of antibody memory to viruses and vaccines. PLoS Biol **2018**; 16:e2006601.
41. Sowers SB, Rota JS, Hickman CJ, et al. High concentrations of measles neutralizing antibodies and high-avidity measles IgG accurately identify measles reinfection cases. Clin Vaccine Immunol **2016**; 23:707–16.
42. 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**; 53:925–33.e4.