

REPORT

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Seroprevalence of SARS-CoV-2 in the Norwegian population measured in residual sera collected in late summer 2020

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Summary

COVID-19 is an infectious disease caused by the novel coronavirus SARS-CoV-2. Infection with SARS-CoV-2 induces antibodies to the virus, therefore the presence of these antibodies in a person's blood indicates that the person has been infected with SARS-CoV-2. This is the second study measuring antibodies against SARS-CoV-2 in residual serum samples collected systematically from various geographical regions in Norway and covering all age groups. The first study with residual sera from April/May 2020 and August 2019, was published as a report in June 2020 (1). In the present study, a total of 1812 residual sera were sampled from 16 microbiology laboratories from late July to mid-September 2020 (week numbers 30-37). The sera were tested for antibodies against SARS-CoV-2 using a novel in-house assay established at the Department of Immunology, Oslo University Hospital. Based on these measurements, the estimated seroprevalence in the Norwegian population in the late summer was 0.6% [95% credible interval (CrI) 0.2 - 1.2]. This is not significantly different from the seroprevalence estimate based on the 900 samples collected in the spring (1.0% [95% CrI 0.1- 2.4])(1). Antibodies against SARS-CoV-2 were found in samples from 6 of the 11 counties, but there were no significant differences between the seroprevalence estimates for each county. However, the sample size from each county varied considerably (from 20 to 387) and the number of positive samples were limited (11 in total). There were no significant differences between the seroprevalence estimates for the various age groups or between males and females. During the summer of 2020, the pandemic was quite contained in Norway. This may explain why the estimated seroprevalence for the Norwegian population was similar in the spring and in the late summer. In this first follow-up study, more laboratories contributed, and a greater number of samples was tested, leading to better precision of the seroprevalence estimate. New follow-up studies of antibodies against SARS-CoV-2 in future collections of residual sera will provide further information about the development of the COVID-19 pandemic in Norway.

Norsk sammendrag

Covid-19 er en infeksjonssykdom som skyldes det nye koronaviruset SARS-CoV-2. Infeksjon med SARS-CoV-2 induserer antistoffer mot viruset, og funn av spesifikke antistoffer mot SARS-CoV-2 i blod indikerer gjennomgått infeksjon med SARS-CoV-2. Dette er den andre studien som måler antistoffer mot SARS-CoV-2 i serumprøver som er systematisk samlet inn fra ulike geografiske områder i Norge, og som representerer alle aldersgrupper. Til sammen 1812 restsera fra 16 laboratorier ble samlet inn i fra slutten av juli til midten av september 2020 (uke 30-37). Serumprøvene ble testet for antistoffer mot SARS-CoV-2 ved hjelp av en ny metode som er utviklet og etablert ved Avdeling for immunologi og transfusjonsmedisin ved Oslo universitetssykehus. Ut ifra disse nye målingene, lå den estimerte andelen av den norske befolkningen som har antistoffer mot SARS-CoV-2 (seroprevalensen) på 0,6 % [95 % kredibilitetsintervall 0,2 – 1,2] i denne perioden. Dette er ikke signifikant forskjellig fra den estimerte seroprevalensen basert på målinger i 900 restsera samlet inn i april/mai 2020 (1,0 % [95 % kredibilitetsintervall 0,1 - 2,4]) (1). Antistoffer mot SARS-CoV-2 ble funnet i prøver fra 6 av de 11 fylkene, men det var ingen signifikante forskjeller i estimert seroprevalens mellom de ulike fylkene. Imidlertid var det stor variasjon i antall prøver fra hvert fylke (fra 20 til 387) og det totale antallet positive prøver var lavt (11 stk). Det ble ikke funnet noen signifikante forskjeller i estimert seroprevalens mellom ulike aldersgrupper eller mellom ulike kjønn. Sommeren 2020 var det lite spredning av SARS-CoV-2 i Norge. Dette kan forklare hvorfor den estimerte seroprevalensen for Norges befolkning holdt seg tilnærmet uendret fra våren til sensommeren. Det var flere laboratorier som bidro og flere restsera som ble testet i denne andre

innsamlingsrunden, slik at det er bedre presisjon i seroprevalensestimater i denne andre studien av seroprevalens i Norge. Nye oppfølgingsstudier av antistoffer mot SARS-CoV-2 i fremtidige innsamlinger av restsera kan gi ytterligere informasjon om utviklingen av covid-19 pandemien i Norge.

Background

A new infectious disease, Coronavirus disease 2019 (COVID-19) was first reported in China in December 2019 and spread rapidly across the world. On March 11th, 2020, the World Health Organization (WHO) stated that COVID-19 constitutes a pandemic (2). COVID-19 is caused by Severe Acute Respiratory Syndrome Corona Virus 2 (SARS-CoV-2), a novel virus belonging to the coronavirus family.

Infection with SARS-CoV-2 induces antibodies to the virus, therefore the presence of these antibodies in a person's blood indicates that the person has been infected with SARS-CoV-2. Some studies have shown that after two weeks of infection, more than 90% of individuals infected with SARS-CoV-2 will have developed antibodies to the virus (3, 4). Seroprevalence is the percentage of a population with antibodies against a certain infectious agent based on analyses in serum or plasma samples. From measurements of the seroprevalence of SARS-CoV-2 in a smaller population, it is possible to estimate the seroprevalence in the Norwegian population.

In Norway, the first COVID-19 case was confirmed on February 26th, 2020. Subsequently, there was a rise in confirmed cases in Norway with a peak incidence on March 26th, 2020 (week number 13), followed by diminishing numbers towards and during the summer as shown in Figure 1 (5, 6). We estimated the seroprevalence of SARS-CoV-2 in the Norwegian population to be close to 1% in the spring of 2020 (1.0% [95% Credible Interval 0.1 %- 2.4%]), based on antibody measurements in 900 residual sera collected between mid-April to mid-May 2020 (weeks 17-20)(1).

During the summer of 2020, the pandemic was quite contained in Norway, but in the late summer of 2020 the numbers of PCR-confirmed positive cases again started rising, representing a second wave of the pandemic (7). By mid-September (week number 37), Norway had 12,287 PCR-confirmed cases of COVID-19 (229 cases per 100,000), and 265 COVID-19-associated deaths (4.9 per 100,000) (8). Between week number 20 and week number 30, the time-period between the two studies, 874 people tested positive. We here present the results of a follow-up study of SARS-CoV-2 seroprevalence in the Norwegian population in the late summer of 2020 based on antibody measurements in residual sera collected between late July to mid-September (weeks 30-37).

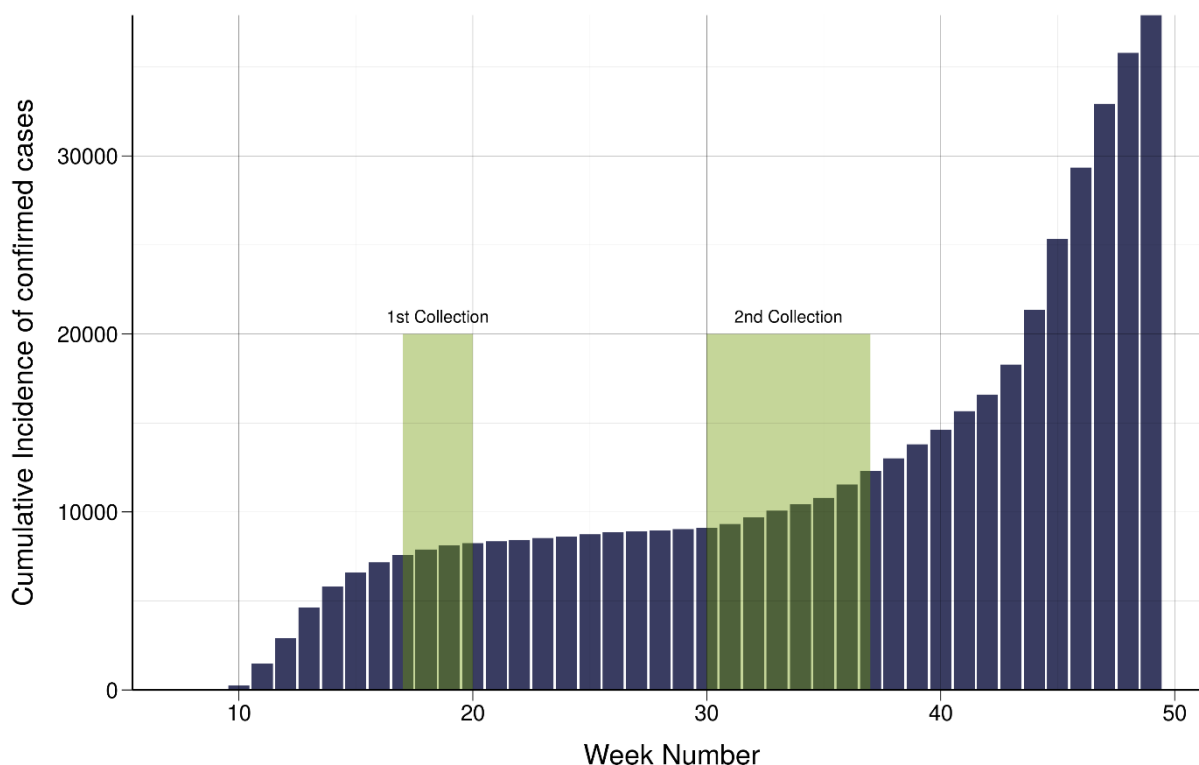


Figure 1. Cumulative incidence of PCR-confirmed COVID-19-cases in Norway by week number as reported to the Norwegian Surveillance System for Communicable Diseases (MSIS). The first collection of residual serum samples occurred between weeks 17-20 (1) and the second collection between weeks 30-37.

Methods

Panels of anonymised residual serum samples were solicited according to a scheme for annual collection that has been operated since the late 1970s as part of serosurveillance of influenza in Norway (9). In order to study the exposure to SARS-CoV-2 at a population level, 1812 residual serum samples were collected from 16 microbiological laboratories across Norway over eight weeks from late July to mid-September 2020 (between week numbers 30-37, i.e. from the 20th of July to the 13th of September). The age of participants was grouped as follows: 0-4, 5-14, 15-24, 25-59 and ≥ 60 years of age. The laboratories were asked to not include sera from individuals with a known HIV, hepatitis B or C diagnosis, or very turbid or haemolysed serum samples.

The sera were analysed using an in-house flow cytometer-based method detecting IgG antibodies against SARS-CoV-2 derived antigens (10). The estimated sensitivity of the test is 86% (95% Credible Interval (CrI) 82%-90%), based on the detection of antibodies against SARS-CoV-2 in serum samples collected at least three weeks from onset of symptoms from individuals with PCR-confirmed COVID-19. The estimated specificity of the test is 99.9% (95% CrI 99.7%-100.0%), based on analysis of 1343 sera sampled before the pandemic. Test sensitivity and specificity have been used to convert proportions of test positives into estimated seroprevalences.

95% confidence interval (CI) for fraction positives were estimated using the exact method. Seroprevalence was estimated for Norway and by laboratory, county of residence, sex, age group and sampling week for subgroups with more than 30 sera. Sera lacking information on county of residence were provisionally attributed to the county corresponding to the laboratory. For the estimation, we used a Bayesian method that incorporates the uncertainties in the sensitivity and the specificity of the test (11). We adjusted the overall seroprevalence by a multilevel regression and poststratification on counties, age groups and sex (11). For seroprevalence results we present a point estimate and a 95% CrI.

Results

Sixteen microbiological laboratories across the country contributed with approximately 113 sera each (range 100-120). Of the overall 1812 samples tested, antibodies against SARS-CoV-2 were detected in 11 samples (0.6%, 95% CI 0.3-1.1). This gives an estimated seroprevalence of 0.6% (95% CrI 0.2-1.2) in the Norwegian population when adjusted for the estimated test sensitivity and specificity, as well as for county, age and sex (Table 1).

The results for samples from the different laboratories in Norway are shown in Table 1. Ten of the 16 contributing laboratories did not have any samples that tested positive. The highest positivity rate for antibodies against SARS-CoV-2 was found for the laboratory in Skien (3.0%, 95% CI 0.6 - 8.5). However, the laboratory in Skien (Unilabs) is a commercial lab receiving samples from all over Norway, and the positive samples from this lab were from individuals living in Oslo (n=2) and Viken (n=1).

The estimated seroprevalence based on county of residence is presented in Table 2. Positive samples were found in sera from individuals living in six of the 11 counties. Most positive samples (n=3) were found among individuals living in Viken, but Viken was also the county of residence with the largest number of samples tested (21.4% of all samples). There were no significant differences in the estimated seroprevalence between the various counties; however, the number of samples tested from each county varied considerably (range 20-387).

We did not find any differences in seroprevalence by sex (Table 3) or age group (Table 4). The residual sera were from 57.3% males and 42.5% females and came from individuals aged 0 to 97 years. The youngest person for whom the serum tested positive was 5 years old, while the oldest was 78 years old. Positive samples were found for all age groups, except for the 0-4 years old category (6.2% of the samples).

The residual sera were collected between week numbers 30-37, i.e. from the 20th of July to the 13th of September 2020 (range 36-821 samples collected per week) (Table 5). Most samples were collected in week number 31 (27th of July – 2nd of August) (45.3%), and 79.5% of the samples were collected in the first three weeks. For 40 (2.2%) of the samples collected between weeks 30-37, information about the specific sampling week was missing. There were no differences between the seroprevalence estimates for the different weeks, but again, most of the credible intervals were wide.

Tables

Table 1. Estimated seroprevalence, overall, and seropositivity rates by submitting laboratory

	Positive samples	Number of samples tested (% of all)	Percent positive samples (95% CI)	Estimated seroprevalence [%] (95% credible interval)
Total	11	1812 (100%)	0.6 (0.3 - 1.1)	0.6 (0.2 – 1.2)*
By submitting laboratory				
Skien**	3	100 (5.5%)	3.0 (0.6 - 8.5)	4.2 (1.2 - 9.8)
Molde	2	116 (6.4%)	1.7 (0.2 - 6.1)	2.6 (0.5 - 7.0)
Drammen	2	111 (6.1%)	1.8 (0.2 - 6.4)	2.7 (0.5 - 7.3)
Trondheim	1	120 (6.6%)	0.8 (0.0 - 4.6)	1.5 (0.2 - 5.2)
Tromsø	2	102 (5.6%)	2.0 (0.2 - 6.9)	2.9 (0.6 - 7.9)
Lillehammer	1	109 (6.0%)	0.9 (0.0 - 5.0)	1.7 (0.2 - 5.8)
Hammerfest	0	119 (6.6%)	0.0 (0.0 - 3.1)	0.7 (0.0 - 3.5)
Ullevål	0	112 (6.2%)	0.0 (0.0 - 3.2)	0.7 (0.0 - 3.6)
Stavanger	0	120 (6.6%)	0.0 (0.0 - 3.0)	0.7 (0.0 - 3.5)
Levanger	0	120 (6.6%)	0.0 (0.0 - 3.0)	0.7 (0.0 - 3.4)
Fredrikstad	0	111 (6.1%)	0.0 (0.0 - 3.3)	0.7 (0.0 - 3.9)
Førde	0	120 (6.6%)	0.0 (0.0 - 3.0)	0.6 (0.0 - 3.5)
AHUS	0	117 (6.5%)	0.0 (0.0 - 3.1)	0.7 (0.0 - 3.5)
Kristiansand	0	106 (5.8%)	0.0 (0.0 - 3.4)	0.7 (0.0 - 4.1)
Bergen	0	120 (6.6%)	0.0 (0.0 - 3.0)	0.7 (0.0 - 3.5)
Bodø	0	109 (6.0%)	0.0 (0.0 - 3.3)	0.8 (0.0 - 3.9)

*The overall seroprevalence for Norway was adjusted for age, sex and county. **All the positive samples from the laboratory in Skien were from persons residing in other counties than Vestfold og Telemark; two were from Oslo and one from Viken (see also Table 2).

Table 2. Estimated seroprevalence by county of residence

County of residence	Positive samples	Number of samples tested (% of all)	Percent positive samples (95% CI)	Estimated seroprevalence [%] (95% credible interval)
Oslo*	2	128 (7.1%)	1.6 (0.2 - 5.5)	2.3 (0.4 - 6.1)
Rogaland	0	143 (7.9%)	0.0 (0.0 - 2.5)	0.5 (0.0 - 3.0)
Møre og Romsdal*	2	135 (7.5%)	1.5 (0.2 - 5.2)	2.2 (0.4 - 5.9)
Nordland	0	115 (6.3%)	0.0 (0.0 - 3.2)	0.6 (0.0 - 3.7)
Viken	3	387 (21.4%)	0.8 (0.2 - 2.2)	1.0 (0.2 - 2.6)
Innlandet	1	117 (6.5%)	0.9 (0.0 - 4.7)	1.6 (0.2 - 5.4)
Vestfold og Telemark	0	20 (1.1%)	0.0 (0.0 - 16.8)	n.a.**
Agder	0	110 (6.1%)	0.0 (0.0 - 3.3)	0.7 (0.0 - 3.9)
Vestland	0	222 (12.3%)	0.0 (0.0 - 1.6)	0.4 (0.0 - 2.0)
Trøndelag	1	218 (12.0%)	0.5 (0.0 - 2.5)	0.8 (0.1 - 2.9)
Troms og Finnmark	2	217 (12.0%)	0.9 (0.1 - 3.3)	1.4 (0.3 - 3.9)

*40 sera from the laboratory in Molde and one of the sera from Oslo (Ullevål) were attributed to the corresponding counties (Møre og Romsdal and Oslo, respectively), as these sera did not include information on county of residence. **n.a.: not applicable. The seroprevalence was not estimated for subgroups with less than 30 samples tested.

Table 3. Estimated seroprevalence by sex

Sex	Positive samples	Number of samples tested (% of all)	Percent positive samples (95% CI)	Estimated seroprevalence [%] (95% credible interval)
Male	5	1039 (57.3%)	0.5 (0.2 - 1.1)	0.6 (0.2 - 1.3)
Female	6	770 (42.5%)	0.8 (0.3 - 1.7)	0.9 (0.3 - 1.9)
Missing	0	3 (0.2%)	0.0 (0.0 - 70.8)	n.a.*

*n.a.: not applicable. The seroprevalence was not estimated for subgroups with less than 30 samples tested.

Table 4. Estimated seroprevalence by age

Age groups (years)	Positive samples	Number of samples tested (% of all)	Percent positive samples (95% CI)	Estimated seroprevalence [%] (95% credible interval)
0-4	0	112 (6.2%)	0.0 (0.0 - 3.2)	0.7 (0.0 - 3.8)
5-14	3	312 (17.2%)	1.0 (0.2 - 2.8)	1.3 (0.3 - 3.2)
15-24	3	308 (17.0%)	1.0 (0.2 - 2.8)	1.3 (0.3 - 3.2)
25-59	2	700 (38.6%)	0.3 (0.0 - 1.0)	0.4 (0.0 - 1.2)
≥60	3	379 (20.9%)	0.8 (0.2 - 2.3)	1.0 (0.2 - 2.6)
Missing	0	1 (0.1%)	0.0 (0.0 - 97.5)	n.a.*

*n.a.: not applicable. The seroprevalence was not estimated for subgroups with less than 30 samples tested.

Table 5. Estimated seroprevalence by sampling week

Sampling week number	Positive samples	Number of samples tested (% of all)	Percent positive samples (95% CI)	Estimated seroprevalence [%] (95% credible interval)
30	2	294 (16.2%)	0.7 (0.1 - 2.4)	1.0 (0.2 - 2.6)
31	3	821 (45.3%)	0.4 (0.1 - 1.1)	0.4 (0.1 - 1.2)
32	2	325 (17.9%)	0.6 (0.1 - 2.2)	0.9 (0.2 - 2.5)
33	2	118 (6.5%)	1.7 (0.2 - 6.0)	2.5 (0.6 - 6.9)
34	0	72 (4.0%)	0.0 (0.0 - 5.0)	1.1 (0.0 - 5.7)
35	1	36 (2.0%)	2.8 (0.1 - 14.5)	5.0 (0.6 - 16.3)
36	0	39 (2.2%)	0.0 (0.0 - 9.0)	2.0 (0.1 - 9.6)
37	1	67 (3.7%)	1.5 (0.0 - 8.0)	2.8 (0.4 - 9.0)
Unknown (within 30-37)	0	40 (2.2%)	0.0 (0.0 - 8.8)	n.a.*

*n.a.: not applicable.

Discussion

This is the second study measuring antibodies against SARS-CoV-2 in residual sera sampled from various geographical regions and covering all age groups in Norway, and a follow-up to a similar study from the spring of 2020 (1). The number of residual serum samples with detected antibodies against SARS-CoV-2 was low, indicating an estimated seroprevalence of 0.6% (95% CrI 0.2 - 1.2) in the Norwegian population by late summer 2020. We did not see an increase in the seroprevalence from the first study based on sera collected in April-May 2020 (1.0% [95% CrI 0.1- 2.4]) to the second sampling round. In this second study, more laboratories contributed,

and the total number of samples was doubled; therefore, the second seroprevalence estimate is more representative for the whole of Norway and with better precision.

After an initial peak in the number of PCR-positive cases of COVID-19 in the spring of 2020, the numbers declined due to the strict non-pharmaceutical interventions taken by the Norwegian government (12). On the 12th of March and in the following days, the Norwegian government issued a partial “lock down” with school and universities closings, closing of the borders and of services such as hairdressers and physiotherapists, and people were asked to work from home as much as possible. From May 2020, there was a gradual re-opening of the Norwegian society, with schools starting again, the closed-down businesses could reopen, and the borders were open to travels to and from the EEC/EU-region in the summer. There were a few, small outbreaks during the summer and in the late summer of 2020, but the cumulative number of PCR-confirmed cases increased very slowly during this period (Figure 1). Consequently, the low number of samples with antibodies against SARS-CoV-2 found in both studies of residual sera matches the relatively low number of PCR-confirmed cases of COVID-19 in Norway notified to the Norwegian Surveillance System for Communicable Diseases (MSIS).

In the last sampling week in the spring (mid-May), 8,254 PCR-confirmed cases of COVID-19 (0.15% of the Norwegian population) had been notified to MSIS (Figure 1) (5). By the last week of July, when 45.3% of the samples in the second study was collected, the cumulative number of COVID-19 cases had increased to 9,128 cases (0.17% of the population) (13). By the end of the second sampling period, the number of notified cases started to increase more rapidly (Figure 1) (8). However, there is a delay of at least two weeks from the time of infection to the time when antibodies can be detected. Thus, this increase in the number of confirmed cases of COVID-19 at the end of the sampling period is not reflected in the number of residual sera positive for antibodies against SARS-CoV-2 in the present study.

In the beginning of May (week number 18), mathematic modelling suggested that between 0.4% and 0.5% of the Norwegian population had been infected, and by the end of July (week number 31), this estimate had increased only slightly to 0.45%-0.62% (14). These results agree well with the seroprevalence estimate of 0.6% reported in the present study. The data presented here, also corresponds with the proportion of samples positive for antibodies against SARS-CoV-2 among a random sample of participants from The Norwegian Mother, Father and Child Cohort Study (MoBa) living in Oslo (1.3% positive samples of 2439 individuals) (15). In the present study, the percent positive samples among individuals with Oslo as their county of residence, was 1.6% in the same time period.

By the end of August 2020, the pandemic in Norway had lasted approximately 6 months with the highest number of cases in the first wave of the pandemic in March 2020 (7). Some of the samples in the present study may have been from individuals infected early in the pandemic. The duration of antibodies against SARS-CoV-2 after infection is not known. Some studies suggest that the antibodies last long, while other indicate that antibodies wane quickly after infection (16-19). However, studies seem to agree that antibodies against SARS-CoV-2 can be found in most individuals at least 3 months after infection (20). Antibodies against SARS-CoV, a related coronavirus, have been found to be detectable for up to one year (4, 19). Based on the literature on duration of antibodies against SARS-CoV-2 and SARS-CoV, it is more likely that the low number of positive samples in the second study is the result of the overall low cumulative number of COVID-19 cases in Norway, rather than waning of antibodies.

The residual sera were collected between late July to mid-September 2020 (weeks 30-37) with most of the samples from the first three weeks. After an infection with SARS-CoV-2, studies have

found that IgG antibodies against SARS-CoV-2 are generally detected 6-14 days following symptom onset, with antibodies being detected in 90% of individuals after 2 weeks, and 100% after 4 weeks (3). In our study, estimation of the test sensitivity was based on samples from individuals with PCR-confirmed COVID-19, and with at least 3 weeks since symptom onset. We cannot rule out that some residual sera included in the study were collected from individuals recently infected with SARS-CoV-2. These individuals may not have developed antibodies at the time of sample collection but could seroconvert later. This will affect the interpretation of the results, as most of the samples were from week numbers 30-32, and the estimated seroprevalence therefore represents infections that occurred up until approximately 3 weeks prior to sampling (i.e. until week numbers 27-30).

Seroprevalence studies may help to determine the number of cases of COVID-19 in a population, as not all cases are tested and confirmed at the time of infection. However, there are several limitations to these studies (21). Using residual sera to estimate population prevalence could lead to selection bias, as the samples came from medical laboratories, potentially including persons with more morbidity, comorbidities or different risk and health-seeking behaviours. Conversely, testing of sera from invited persons may lead to healthy-person biases or non-participation of certain groups (22). However, the consistent procedure of sampling of residual sera in the first and the second seroprevalence studies, makes the findings comparable over time. The level of antibodies against SARS-CoV-2 has been reported to be linked with severity, where individuals who are asymptomatic or have mild COVID-19 may have lower levels of antibodies than individuals with severe disease (4, 17). If these less severe cases are not detected in antibody assays due to low levels of antibodies, this can lead to underestimation of the seroprevalence. Furthermore, it is possible that not all individuals infected with COVID-19 will mount an antibody response. In the present study, only IgG antibodies were measured and used to estimate the seroprevalence of SARS-CoV-2 in the Norwegian population (21). SARS-CoV-2 infects the respiratory tract, and IgA antibodies induced by infection may also be relevant for protection against COVID-19. The inclusion of analyses of IgA and as well as IgM antibodies against SARS-CoV-2 may increase the sensitivity of serological testing (21).

Conclusion

The estimated seroprevalence in the overall Norwegian population was still low in the late summer of 2020 (0.6% [95% CrI 0.2 - 1.2]), and not significantly different from the estimated seroprevalence in the spring of 2020. This finding fits well with the low number of PCR-detected cases of COVID-19 during the summer when the pandemic was quite contained in Norway. Measurements of antibodies against SARS-CoV-2 in future collections of residual sera may provide further information about development of the COVID-19 pandemic in Norway, especially as the number of PCR-detected cases started rising again from September 2020.

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