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RESEARCH ARTICLE

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Severe acute respiratory syndrome coronavirus 2 prevalence in 1170 asymptomatic Norwegian conscripts

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Abstract

Background: Accurate estimates of SARS-CoV-2 infection in different population groups are important for the health authorities. In Norway, public infection control measures have successfully curbed the pandemic. However, military training and service are incompatible with these measures; therefore extended infection control measures were implemented in the Norwegian Armed Forces. We aimed to describe these measures, discuss their value, and investigate the polymerase chain reaction (PCR) prevalence and seroprevalence of SARS-CoV-2, as well as changes in antibody titer levels over the 6-week military training period in a young, asymptomatic population of conscripts.

Methods: In April 2020, 1170 healthy conscripts (median age 20 years) enrolled in military training. Extended infection control measures included a pre-enrollment telephone interview, self-imposed quarantine, questionnaires, and serial SARS-CoV-2 testing. At enrollment, questionnaires were used to collect information on symptoms, and SARS-CoV-2 rapid antibody testing was conducted. Serial SARS-CoV-2 PCR and serology testing were used to estimate the prevalence of confirmed SARS-CoV-2 and monitor titer levels at enrollment, and 3 and 6 weeks thereafter.

Results: At enrollment, only 0.2% of conscripts were SARS-CoV-2 PCR-positive, and seroprevalence was 0.6%. Serological titer levels increased nearly 5-fold over the 6-week observation period. Eighteen conscripts reported mild respiratory symptoms during the 2 weeks prior to enrollment (all were PCR-negative; one was serology-positive), whereas 17 conscripts reported respiratory symptoms and nine had fever at enrollment (all were PCR- and serology-negative).

Conclusions: The prevalence of SARS-CoV-2 was less than 1% in our sample of healthy Norwegian conscripts. Testing of asymptomatic conscripts seems of no value in times of low COVID-19 prevalence. SARS-CoV-2 antibody titer levels increased substantially over time in conscripts with mild symptoms.

KEYWORDS
adolescents, Armed Forces, conscripts, coronavirus, COVID-19, prevalence, SARS-CoV-2, serological analyses, serology, youths

1 | INTRODUCTION

Current knowledge suggests that children and young adults infected with SARS-CoV-2 are more often asymptomatic, or have fewer and milder symptoms than older patients.[1-3] Moreover, children and teens between 10 and 19 years of age may be more likely to spread the virus among family members than adults and younger children.[4] Polymerase chain reaction (PCR) testing is mainly performed on symptomatic patients, those in need of hospitalization, patients at risk, and among health care workers. So far, SARS-CoV-2 testing has not been performed systematically in asymptomatic groups, thus the prevalence and rate of transmission in young asymptomatic individuals are still largely unknown. Accurate estimates of infection within different population groups are crucial for health authorities when deciding how and when to close and reopen societies during the COVID-19 pandemic.

Military service in Norway is mandatory for all men and women; between 7000 and 8000 of them undergo military conscription every year.[5] In order to ensure redundancy, enrollment of new conscripts is a continuous process in the Norwegian Armed Forces. In Norway, public infection control measures have successfully curbed the rates of hospital admissions and deaths due to COVID-19.[6] However, military training and service are not compatible with these measures, as service personnel live together in barracks, and combat training involves body contact. Therefore, in mid-April 2020, the Norwegian Armed Forces implemented extended infection control measures to ensure the continuation of military training and the health of military personnel. The measures included a pre-enrollment telephone interview, self-imposed quarantine before enrollment, questionnaires, and serial SARS-CoV-2 PCR and serology testing during the initial 6-week training period. We aimed to describe these measures, discuss their value, and investigate the PCR prevalence and seroprevalence of SARS-CoV-2, as well as changes in antibody titer levels over the 6-week military training period in a young, asymptomatic population of conscripts.

2 | MATERIALS AND METHODS

2.1 | Cohort

The study cohort included 1170 healthy conscripts (median age: 20 years, range 18-25), 798 men (68.2%) and 372 women (31.8%), who enrolled in military training between 19 and April 27, 2020. As conscripts are called for military service regardless of residential area, our cohort included men and women from all over Norway.

2.2 | Extended infection control measures

Two weeks prior to enrollment, conscripts were interviewed by telephone to motivate them for military service and ensure compliance with public infection control measures related to COVID-19. All conscripts were encouraged to undergo self-imposed quarantine until enrollment.

On enrollment day, conscripts underwent an initial screening: they were asked about current respiratory symptoms, their body temperature was measured using an ear thermometer, and a sample was taken for rapid antibody testing (performed on site). Individuals with symptoms, a temperature ≥38.0°C, and/or a positive rapid antibody test underwent further clinical interviews and examinations, and were then quarantined pending the results of a PCR test (Figure 1).

All conscripts also completed an online questionnaire, which collected information on possible or confirmed COVID-19 over the previous 2 weeks, compliance with public infection control measures, and possible close contact with SARS-CoV-2-infected individuals.

2.3 | SARS-CoV-2 rapid antibody test, PCR, and serology analyses

Capillary blood samples were used for rapid antibody testing (Acro 2019-nCoV IgG/IgM, Acro Biotech Inc., Rancho Cucamonga, California). This test is a lateral flow chromatographic immunoassay for qualitative detection of IgG and IgM antibodies to SARS-CoV-2.

Nasopharyngeal swabs were used to collect samples for SARS-CoV-2 PCR at enrollment, and after 3 and 6 weeks of military training. PCR tests (Roche cobas SARS-CoV-2 RNA test, Roche Diagnostics, Hoffmann-La Roche, Basel, Switzerland) were run at the Department of Microbiology at Oslo University Hospital on the Cobas 6800 platform. The cobas SARS-CoV-2 RNA test is a single-well dual target assay, which includes both specific detection of SARS-CoV-2 and pan-Sarbecovirus detection for the Sarbecovirus subgenus family that includes SARS-CoV-2.[7]

Venous blood samples were collected using VACUETTE Blood Collection Tubes (Greiner Bio-One, Kremsmünster, Austria) for

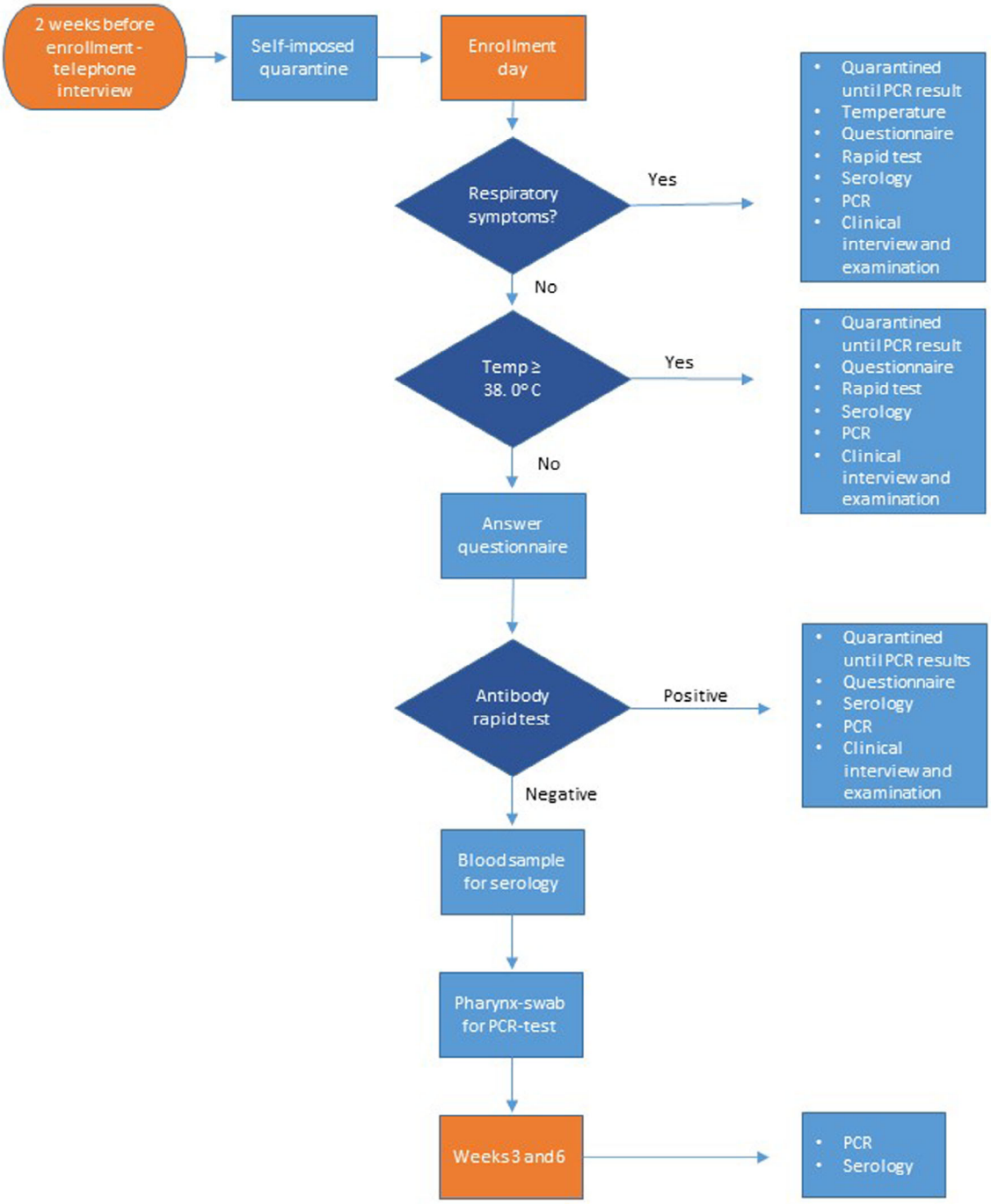

FIGURE 1 Extended infection control measures in the Norwegian Armed Forces

SARS-CoV-2 serology testing at enrollment, and after 3 and 6 weeks of military training. Samples were analyzed at the Department of Microbiology at Oslo University Hospital, using the Elecsys anti-SARS-CoV-2 IgM/IgG assay fully automated on the Cobas e801 analyzer (Roche Diagnostics). The assay measures the combined total of IgM and IgG against the nucleocapsid (N) structural protein of SARS-CoV-2 and provides results as a cutoff index (COI) calculated based on signal sample/assay cutoff. A COI above 1.2 was defined as a positive result.

2.4 | Ethics

Data resulting from the implementation of extended infection control measures in the Norwegian Armed Forces are administered by the Norwegian Armed Forces Health Registry (NAFHR), a central health registry with data from Norwegian Armed Forces personnel, including conscripts, and civilian and military staff. Current regulations authorize the NAFHR to produce anonymous statistics for research purposes.[8]

3 | RESULTS

On enrollment day, 17 (1.4%) of the 1170 conscripts reported mild respiratory symptoms, and nine (0.8%) had a body temperature ≥38.0°C. None of these conscripts was SARS-CoV-2 PCR-positive, or serology-positive at enrollment.

Two of the 1170 conscripts were both SARS-CoV-2 rapid antibody test IgG- and IgM-positive, 25 were IgM-positive only, and 29 were IgG-positive only. Of the 1170 conscripts, seven were both SARS-CoV-2 rapid antibody test IgG-positive and serology-positive (Table 1), while none were rapid antibody test-negative and serology-positive. Using the serological test at Oslo University Hospital as the gold standard, the sensitivity of the SARS-CoV-2 rapid antibody test was calculated to be 100%. Of the 56 conscripts who were SARS-CoV-2 rapid antibody test-positive, 49 were serology-negative, whereas 1114 were both serology-negative and rapid antibody test-negative. Therefore, a total of 1163 were serology-negative, and the specificity of the SARS-CoV-2 rapid antibody test was calculated to be 96%. In this cohort, which had a seroprevalence at enrollment of 0.6%, the positive predictive value of the rapid antibody test was calculated to be 13%.

In the online questionnaire, 18 conscripts (1.5%) reported mild respiratory symptoms during the 2 weeks prior to enrollment. Reported symptoms included runny nose and sneezing (72%), stuffy nose (61%), cough (55%), sore throat (33%), headaches (33%), shortness of breath (16%), fever (13%), reduced sense of taste or smell (11%), sore muscles (11%), and dizziness (6%). None of these 18 conscripts reported any respiratory symptoms on enrollment day. None was SARS CoV-2 PCR-positive at enrollment, but one was serology-positive. Nine conscripts (0.8%) answered "yes" to the question "Do you think you have had COVID-19?". Among these, one was SARS-CoV-2 PCR-positive and four were serology-positive at enrollment.

Two of the 1170 conscripts were SARS-CoV-2 PCR-positive at enrollment, and one conscript who was initially PCR-negative tested positive at week 6 (Table 1). None of these individuals reported any symptoms during the 2 weeks prior to enrollment, at enrollment, or during the observation period. However, during additional clinical interviews, the two conscripts who were PCR-positive at enrollment reported illness with symptom onset 4 and 7 weeks prior to enrollment, respectively. The individual who tested positive at week 6 had not been ill, and experienced no symptoms before or during the 6-week observation period.

During the 6-week observation period, a total of eight conscripts were SARS CoV-2 serology-positive, one female, and seven males. Seven conscripts (0.6%) were SARS-CoV-2 serology-positive at enrollment (Table 1), including the three who were also PCR-positive, and one was SARS-CoV-2 PCR-negative and serology-negative at enrollment, but developed positive and rising titers at weeks 3 and 6. None of these eight conscripts had fever or reported respiratory symptoms at enrollment, nor did they report any respiratory symptoms during the observation period. One reported mild respiratory symptoms during the 2 weeks prior to enrollment. However, based on the additional clinical interview, six of the eight SARS-CoV-2 serology-positive conscripts had been ill with symptom onset 3 to 7 weeks before enrollment (Table 1). Antibody titers increased almost 5-fold during the 6-week observation period in all but one of the eight SARS-CoV-2 serology-positive conscripts (Table 1 and Figure 2).

TABLE 1 SARS-CoV-2 rapid antibody test, PCR, and serology results at enrollment (W0), week 3 (W3), and week 6 (W6), number of days between symptom onset and W0, symptom description, and disease duration for the eight conscripts with positive serology during the observation period

ID	Rapid-antibody test IgG	PCR W0	PCR W3	PCR W6	Sero COI[a] W0	Sero COI[a] W3	Sero COI[a] W6	Days between symptom onset and W0	Symptoms	Disease duration
1	pos	neg	neg	neg	57.4	66.0	51.7	48 days	Dry cough	2 days
2	pos	neg	neg	neg	19.6	24.3	27.0	33 days	Headache, fatigue, dry cough, stuffy nose, nausea	5 days
3	pos	neg	neg	neg	19.2	67.8	92.3	22 days	Loss of taste and sense of smell, a little headache	10 days
4	pos	pos	neg	neg	19.2	62.1	89.1	30 days	Sore throat, cough, headache, heavy breathing during activity, loss of taste and sense of smell	10 days
5	pos	neg	neg	pos	18.7	20.3	23.2	Not ill	No symptoms	
6	pos	pos	neg	neg	16.2	40.4	52.0	49 days	Stuffy nose, slight cough, reduced taste	5 days
7	pos	neg	neg	neg	2.1	2.9	7.4	39 days	Fever, fatigue, runny nose, headache, chest pain, muscle pain	10 days
8	neg	neg	neg	neg	0.6	1.5	1.6	Not ill	No symptoms	

Abbreviation: COI, cutoff index.
[a]COI for positive test = 1.2.

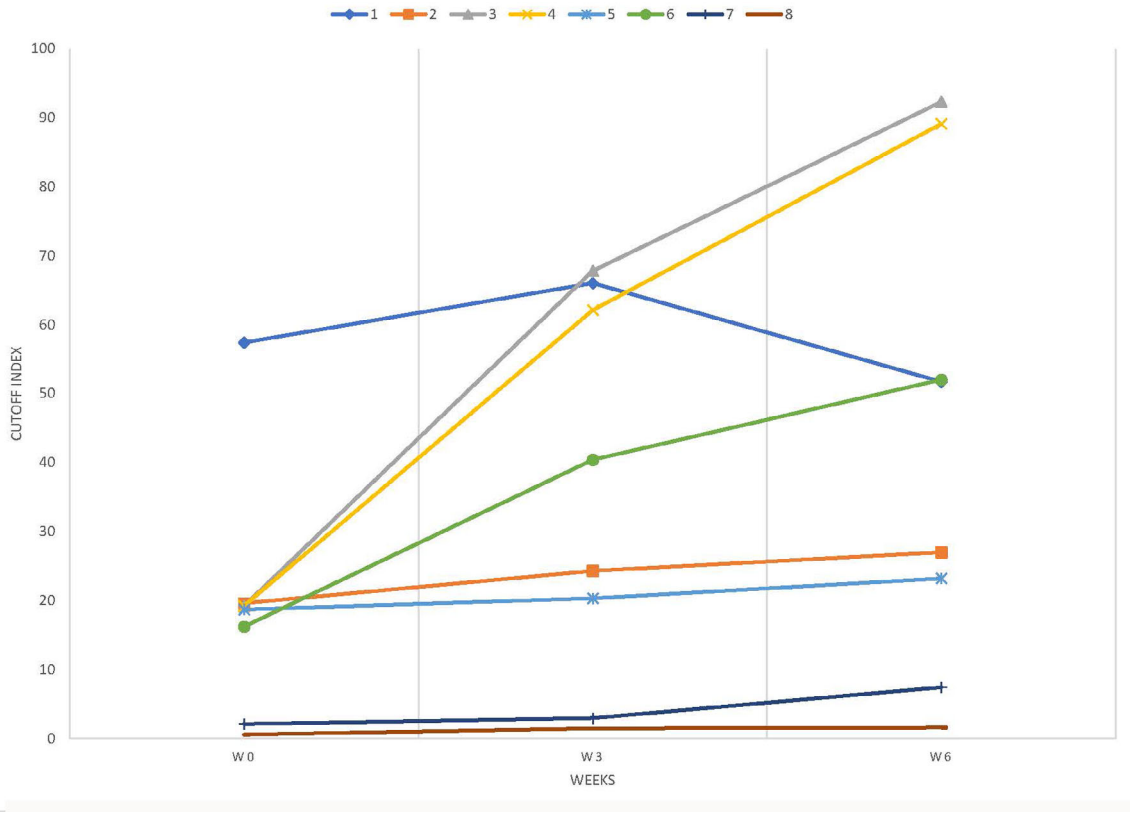

FIGURE 2 Titers of SARS CoV-2 IgG antibodies at enrollment (W0), week 3 (W3), and week 6 (W6) for the eight conscripts with positive serology during the observation period. ID numbers correspond to Table 1

4 | DISCUSSION

In this study of 1170 healthy conscripts, the seroprevalence of SARS-CoV-2 was 0.6%, and 0.2% were SARS-CoV-2 PCR-positive on the day of enrollment to military service. Eighteen conscripts reported mild respiratory symptoms during the 2 weeks prior to enrollment (all were PCR-negative, one was serology-positive), whereas 17 conscripts reported respiratory symptoms and nine had fever at enrollment (all were PCR- and serology-negative). These low numbers may partly be due to the high awareness of public infection control measures in Norway during the observation period, which was 3 to 4 weeks after the pandemic peaked in the country at the end of March/beginning of April 2020.

4.1 | SARS-CoV-2 rapid antibody test, PCR, and serology analyses

The sensitivity and specificity of the rapid antibody test used in this study were 100% and 96%, respectively. The relatively large proportion of false-positive rapid antibody tests is probably partly due to the fact that the manufacturer's instructions recommended that "*The intensity of the color in the test line regions may vary depending on the concentration of SARS-CoV-2 antibodies present in the specimen. Therefore, any shade of color in the test line region should be considered positive.*"[9] This may have resulted in negative tests being interpreted as positive based on nearly invisible lines in the test field, and illustrates that adequate instruction and training of health personnel is crucial when introducing and using new rapid tests. The Acro rapid antibody test has recently been evaluated by the Norwegian Organization for Quality Improvement of Laboratory Examinations and was rated "acceptable," with a sensitivity of 0.88 and a specificity of 0.99.[10]

Two conscripts were SARS-CoV-2 PCR-positive at enrollment, and one tested PCR-positive after 6 weeks. Whether this represented ongoing or previous subclinical infection is unclear. PCR assays detect the presence of viral RNA. Whether positive SARS-CoV-2 PCR represents viable and contagious virus, or only viral remnants, does vary depending on patients and stages of disease. One study found that infectivity peaked 3 days after symptom onset, and the authors were not able to culture the virus from samples obtained more than 8 days after symptom onset.[11]

Eight of the 1170 conscripts (0.7%) were seropositive for SARS CoV-2 IgG during the observation period. As antibodies against SARS-CoV-2 can be detected in the middle and later stages of the COVID-19 illness,[12] testing may help confirm COVID-19 diagnoses in individuals who have not previously been referred to PCR testing. Systematic antibody testing may also shed light on the actual prevalence of COVID-19 in the general population, and thus their immunization status, which is key to the overall pandemic response in most countries. Recent studies have revealed prevalence numbers in the range of 1%

to 5% in European countries.[13] Our results indicate that the prevalence of COVID-19 among young, asymptomatic adults in Norway is less than 1%, which is in agreement with recent mathematical estimates from the Norwegian Institute of Public Health.[6]

Six of eight seropositive conscripts, among whom two were also SARS-CoV-2 PCR-positive, had a history of symptoms compatible with COVID-19, with symptom onset between 3 and 7 weeks before enrollment. Their titer levels increased nearly 5-fold over the 6-week observation period, but they reported no new symptoms. Most people infected with SARS-CoV-2 display an antibody response between 10 and 14 days after infection; however, the antibody response may depend on disease severity.[14] In some mild cases, antibody detection is only possible long after symptom onset, and in a few cases, antibodies are not detected at all, at least not during the time scale of the reported studies.[15] The duration of antibody response is still unknown, but it is known that antibodies to other coronaviruses wane over time (range: 12-52 weeks from the onset of symptoms).[15] One study found that SARS-CoV-2 IgM and IgG antibody levels may remain for 7 weeks after symptom onset.[16]

The symptoms and disease reported by the aforementioned six seropositive conscripts were mild. COVID-19 is generally considered a mild disease in adolescents.[2,3,17] One of the questions that requires further study is to what extent young COVID-19 patients actually present with no symptoms at all, which may increase the risk of spreading the virus to others. Our study confirms that symptoms are generally mild among young adults, but hardly supports the idea of frequent occurrence of asymptomatic spreaders; at least not in Norway.[1]

4.2 | Value of testing and self-reported data

Testing of asymptomatic conscripts in our study cohort did not identify any individuals suspected of active transmission, as both conscripts who were PCR-positive at enrollment had been symptom-free for more than 20 days when they were tested. Seven of the eight conscripts with a positive serology result during the observation period had been symptom-free for at least 14 days before enrollment.

Testing of symptomatic conscripts, on the other hand, is widely recommended,[18] and is used as both as a diagnostic tool and a screening method to detect cases of COVID-19 in areas with active outbreaks. The incidence numbers in Norway have remained under 5% throughout the pandemic. Symptomatic testing in this study cohort did not confirm SARS-CoV-2 infection in any of the 26 conscripts who reported having mild respiratory symptoms or who had fever at enrollment.

Only one of the eight people with positive serology result during the observation period reported respiratory symptoms during the 14 days before enrollment. Given the reported time of symptom onset among all eight conscripts, five would probably have been contagious if they had been enrolled a month earlier, and three might have presented symptoms of COVID-19. Therefore, self-reported data and questionnaires could be effective tools in the detection of COVID-19, especially in combination with symptom-based testing. Asymptomatic testing does not seem justified, given the low prevalence and corresponding low predictive value of tests in this cohort of young healthy conscripts.

5 | CONCLUSION

The prevalence of SARS-CoV-2 was less than 1% in our sample of healthy Norwegian conscripts. Self-reported symptoms and questionnaires may prove useful in detecting COVID-19, especially in combination with symptom-based testing. Asymptomatic testing seems of no value in times of low COVID-19 prevalence. SARS-CoV-2 antibody titer levels increased substantially over time in young adults with relatively mild symptoms.

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CONFLICT OF INTEREST

None of the authors declares any conflict of interest.

TRANSPARENCY STATEMENT

Einar Kristian Borud declares that the manuscript is an honest, accurate, and transparent account of the study being reported; that no important aspects of the study have been omitted; and that any discrepancies from the study as planned (and, if relevant, registered) have been explained.

AUTHOR CONTRIBUTIONS

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Einar Kristian Borud had full access to all of the data in the study and takes complete responsibility for the integrity of the data and the accuracy of the data analysis.

Author approval: All authors have agreed on the order in which their names are listed in the article.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the Norwegian Armed Forces Health Registry. Restrictions apply to the availability of these data, which were used under license for this study.

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