Cross Sectional Prevalence of SARS-CoV-2 antibodies in Health Care Workers in Paediatric Facilities in Eight Countries

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Title: Cross Sectional Prevalence of SARS-CoV-2 antibodies in Health Care Workers in Paediatric Facilities in Eight Countries

Running title: SARS-CoV-2 antibodies in Health Care Workers

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Summary

Background

Healthcare workers (HCWs) have been disproportionately affected by COVID-19 which may in part be driven by nosocomial exposure. If HCW exposure is predominantly nosocomial, HCWs in paediatric facilities, where few patients are admitted with COVID-19, may lack antibodies to SARS-CoV-2 and be at increased risk during the current resurgence.

Aims: To compare SARS-CoV-2 seroprevalence amongst HCWs in paediatric facilities in seven European countries and South Africa (n=8).

Methods

All categories of paediatric HCWs were invited to participate in the study irrespective of previous symptoms. A single blood sample was taken and data about previous symptoms documented. Serum was shipped to a central laboratory in London where IgG to SARS-CoV-2 was measured.

Findings

4114 HCWs were recruited between 1st May and mid-July 2020. The overall seroprevalence range was 0-16.93%. The highest seroprevalence was in London (16.93%) followed by that in Cape Town, South Africa (10.36%). There were no positive HCWs in the Austrian, Estonian and Latvian cohorts, 2/300 positive in Lithuania (0.66%, 0.18-2.4), 1/124 (0.81%, 0.14-4.3) in Romania, and 1/76 (1.3%, 0.23-7.0) in Greece.

Conclusion

The overall seroprevalence amongst paediatric HCWs is similar to their national populations and linked to national COVID-19 burden. Staff working in paediatric facilities in low burden
countries have very low rates of seroprevalence and thus are likely to be susceptible to COVID-19. Their susceptibility to infection may impact on the ability to provide care in the face of increasing COVID-19 disease and highlights the need for appropriate preventative strategies in paediatric health care settings.

Keywords:
COVID-19
SARS-CoV-2
Seroprevalence
Health Care Workers
Hospital Workers
Introduction

Severe acute respiratory syndrome-related coronavirus-2 (SARS-CoV-2) was first recognised in December 2019 and rapidly spread world-wide with the WHO declaring a COVID-19 pandemic on March 11th, 2020. Soon after the identification and genetic sequencing of the virus, diagnostic tests became available for the detection of live virus in human secretions followed rapidly by tests designed to measure serum antibodies to SARS-CoV-2 antigens. Antibodies to SARS-CoV-2 are known to start increasing within 5 days of exposure[1] and IgG can be detected in the serum for many months following exposure. Sero-epidemiology, the presence of antibody in a representative community sample, can shed light on overall population exposure and, when correlates of protection are better understood, may help predict both individual and community susceptibility to infection.

Healthcare workers (HCWs) have had to continue to work throughout the pandemic and the incidence of COVID-19 among HCWs has been shown, in some studies, to be higher than in the general population.[2, 3] Recently the WHO has indicated that while HCWs represent less than 3% of the population in the large majority of countries and less than 2% in almost all low- and middle-income countries, around 14% of COVID-19 cases reported to WHO are among health workers. In some countries, the proportion can be as high as 35% (https://www.who.int/news-room/detail/17-09-2020-keep-health-workers-safe-to-keep-patients-safe-who accessed 5th October 2020).

Studying seroprevalence in HCWs has been undertaken to help understand the transmission potential of the virus in the context of nosocomial exposure and also for insight into the true burden of underlying COVID-19 infection and has been shown in many setting to be
significantly higher than in the relevant general population.[4-7] As relatively few children globally have been admitted to hospital with COVID-19,[8] staff working in paediatric facilities have been unlikely to face a significant risk of exposure from their patients.[9] Only two studies of seroprevalence in paediatric HCWs have been published to date. In Barcelona, Spain, seroprevalence rates of 4% were discovered,[10] similar to the rate in a population-based random sample from the general population in Barcelona (5.4%) analysed at roughly the same time (March/April 2020) while in Argentina, the rate in Paediatricians from a single children’s hospital 3 months into the pandemic was 0.9%.[11] While SARS-CoV-2 has been intensely studied in high prevalence countries that bore the brunt of the initial pandemic, nothing is known about paediatric HCW serostatus in other countries. Understanding levels of seroprevalence in such staff could provide insights into the general prevalence of SARS-CoV-2 in cities or countries where mass testing for the presence of virus in swabs or community based sero-surveys have not been widely implemented or is suboptimal and thereby uncover rates of asymptomatic infection that have led to sero-conversion. Additionally, such information can help health care facilities plan for the current surge in SARS-CoV-2 infection and where relevant target vaccine delivery.

There is currently little standardisation of assays designed to measure antibodies to SARS-CoV-2 resulting in assays of varying sensitivity and specificity being utilised[12] and a consequent difficulty in comparing seroprevalence rates between studies and/or countries. We therefore designed this study to compare rates of SARS-CoV-2 antibody positivity in HCWs working in paediatric facilities in seven different European countries and South Africa. By centralising all of the testing in a single laboratory in London, UK we eliminated methodological laboratory issues that may influence comparisons. In order to contextualise potential differences in seroprevalence rates between the countries participating in this
study we accessed publicly available data on the dates of the initial cases and subsequent epidemiology of COVID-19 in each participating country, the mobility of citizens in each country relative to national restrictions on movement and the government responses during the pandemic.

Methods

This study was undertaken in HCWs in paediatric facilities in seven European and one South African site. The study was initiated at Great Ormond Street Hospital, London, UK, where local hospital staff were invited to enrol in a prospective longitudinal cohort study of SARS-CoV-Serology (COSTARS, IRAS 282713, ClinicalTrials.gov Identifier: NCT04380896). Collaborators based in paediatric health care facilities were invited to join a multi-centre study with a similar design. Ethics approval was obtained locally by the lead investigators of each site. A material transfer agreement which defined the aims of the study and governed the transfer of data and serum was signed between each centre and the principal investigator’s laboratory at the Great Ormond Street Institute of Child Health, University College London. Staff of all categories in each health care setting were invited to join the study irrespective of symptoms or whether they previously suspected they had COVID-19. The numbers of staff recruited were a convenience sample matched to the capacity for laboratory testing. Staff who consented to join the study provided a single 2ml blood sample and filled out a questionnaire focussed on documenting symptoms of SARS-CoV-2 infection since the onset of the pandemic, prior known exposure and the outcome if a viral swab for SARS-CoV-2 RNA PCR had been obtained.
Serology: Serum was prepared and aliquoted, given a unique identifier locally and stored frozen until batch shipping to the WHO International Reference laboratory for Pneumococcal Serology at University College London, London, UK where samples were analysed for the presence IgG to SARS-CoV-2 nucleocapsid protein (Epitope Diagnostics Inc, San Diego, USA) as previously described.[13] All positive or equivocal samples were re-assayed in a multiplexed assay measuring IgG to SARS-CoV-2 nucleocapsid protein, receptor binding domain of S1 and trimeric spike antigen (MSD® SARS-Coronavirus Plate 1, Rockville, MD) to confirm positivity and to establish whether equivocal samples were positive or negative. The MSD assay has undergone extensive evaluation in the London laboratory.[14]

Data sources: National case counts at the time of sampling were taken from data collated from WHO, CDC, and other sources and available on the public website bing.com (https://www.bing.com/covid). Numbers of cases were converted to rates of COVID-19 infection per 100,000 of the population using population estimates published by Eurostat (https://ec.europa.eu/eurostat/data/database). Mobility data for each individual country in the period between the onset of the first case and cohort recruitment was taken from publicly available Google Mobility Data available at https://www.google.com/COVID-19/mobility/. Government responses to the pandemic were accessed at the publicly available website hosted by the Blavatnik School of Government at the University of Oxford https://www.bsg.ox.ac.uk/research/research-projects/coronavirus-government-response-tracker. The Oxford COVID-19 Government Response Tracker (OxCGRT) systematically collects information on several different common policy responses that governments have taken to respond to the pandemic on 17 indicators such as school closures and travel restrictions.
Results

Cohort Characteristics: A total of 4114 HCWs were recruited between the 1st May and mid-July 2020 from the nine sites although the cohort size varied significantly between the centres (Table I). For four countries, Estonia, Latvia, Lithuania and Romania, more than 90% of the staff included in the study were female while females dominated all of the cohorts reflecting the gender makeup of staff in the paediatric health setting. The mean age of participants in each cohort was similar and ranged from 38 to 50 years of age although cohorts had a wide age range (19 years to 78 years). The time taken to recruit the entire cohort varied between sites. Some recruited staff over days, while others continued recruitment over months. The proportion of workers in each cohort reporting symptoms compatible with COVID-19 prior to recruitment varied significantly from <1% (Greece and Romania), 9-18% (Austria, Estonia, Lithuania), 22% (Latvia) to 30-45% (South Africa and UK).

The majority of cohort recruitment took place between May and June 2020 with the exception of Greece, whose recruitment started in mid-June and extended into July, Austria who recruited only in July and South Africa who recruited from mid-June till mid-August 2020 which coincided with the peak of the pandemic (https://www.nicd.ac.za/diseases-a-z-index/covid-19/surveillance-reports/). London recruited throughout May and June and the two periods have been separated out for analytical purposes.

Despite 16-22% of staff in Austria, Estonia, and Latvia reporting symptoms prior to recruitment to the study, none had a positive viral swab for SARS-CoV-2 RNA recorded. In Austria all staff participating in the study were regularly swabbed (two weekly) as part of local health measures. The proportion of the cohort with a positive PCR in Lithuania,
Romania and the UK was less than 1% and all those with positive PCRs were also antibody-positive. The PCR-positive rate in South Africa was higher at 7.66%.

Serology: Seroprevalence rates for three of the four countries with no positive PCRs was zero although Greece had 1 of 76 workers IgG-positive (Figure 1). Similarly, low seroprevalence was found in Romania who had 1/224 workers IgG-positive (0.8%) and Lithuania where only 2 of 300 staff tested IgG positive (0.66%). The seroprevalence in Cape Town HCWs was 10.4% and 15.4% and 16.93% of the London cohort were IgG-positive for the May and June cohorts respectively. When comparing the seroprevalence for the individual cohorts with the rates of COVID-19 cases/100,000 in each country at the time of sampling, some anomalies were noted (Table 1). For those countries with rates below 100 cases per 100,000 of the population (Greece, Latvia, Lithuania, and Romania) seroprevalence rates were low (0-1.3%). However, Estonia and Austria with rates of 148.23 and 225.76/100,000 respectively had no sero-positive HCWs in their cohorts despite 17% of their cohorts reporting symptoms compatible with COVID-19 (although no PCR-positives). UK and South Africa had high rates of COVID-19 at the time of recruitment and this was reflected in high seroprevalence rates.

The time reported between symptoms compatible with COVID-19 and blood sampling was similar for those cohorts with significant numbers of symptomatic staff. The number of days ranged from 89 (UK, June cohort) to 116 days (Austria) and are unlikely to be responsible for the observed differences between countries. South Africa had the shortest mean time between symptoms and blood sampling (46 days) yet had seroprevalence rates lower than the UK. Furthermore the two UK cohorts had different times between symptoms and sampling (64 days for the May cohort and 89 for the June cohort) but seroprevalence rates
were in fact higher for the cohort with longer gap suggesting that over a period of 60-90 days, waning antibody is unlikely to be relevant.

Mobility and government response: Google mobility data, looking at changes in non-residential activities including visits to retail outlets, parks, driving and use of public transport for the relevant areas of each country in this analysis revealed some differences between the countries but none appeared to correlate with seroprevalence (Table 2). The smallest change for the eight countries was Estonia with a -11% change followed by Latvia and Lithuania with -19 and -21% change respectively, yet these three countries all had seroprevalence of <1%. For the other six countries there was a greater reduction in mobility during the initial phases of the pandemic with changes ranging from -31% (Austria) to -42% (Romania). The countries with the highest seroprevalence showed changes of -40% (South Africa) and -33% (UK).

The Oxford COVID-19 government response tracker score was compared for each country at 100 days following the first case which was approximately the 1st or 2nd week of June 2020 for the eight countries in this study (Table 2). For the countries with low seroprevalence rates (and generally lower case rates) the scores ranged from 50-70%. The country with the highest score was South Africa (90%) which reflects the severe lockdown imposed early on in the pandemic. Despite the high score, the seroprevalence rate in the South African cohort was the second highest in this study suggesting that the score may accurately reflect national responses to the pandemic but the response does not necessarily predict the spread of SARS-CoV-2 in a population.

Discussion
Health care workers (HCWs) have continued to work through the pandemic to maintain health services and have therefore been at increased risk of exposure to SARS-CoV-2. As infection with COVID-19 may be asymptomatic additional methods are required to estimate the true burden of disease and by implication the number of staff that might be protected as a result of recovering from infection. Detection of virus in throat/nasal swabs and serological tests of specific antibody have both been used in studies of clinical facing frontline HCWs to try and quantify the burden of disease in this group. Estimates of seroprevalence amongst HCWs have however varied widely. Within Europe, estimates of seroprevalence rates for HCWs range widely; studies in Germany,[15] Greece,[16] Croatia,[17] and Austria[18] have demonstrated low rates (1%, 1.07%, 2% and 3.2%, respectively) while Belgium (6.4%),[19] Spain (9.3%),[6] and the UK, in some studies,[4, 20] have had higher rates. The interpretation of differences in seroprevalence rates between countries is however complicated by the fact that rates have been shown to vary widely between studies in the same country. For example five independent studies conducted within hospitals that are part of the UK National Health Service have varying seroprevalence estimates; from 10.6%[21] or 10.7%[22] at entry to the study to an overall rate of 24.4%[4] or 31.64%[7] or in one study, 25% at entry with a subsequent overall rate of 45%.[20] In the USA, a convenience sample of frontline HCWs who worked with COVID-19 patients at 13 geographically diverse US academic medical centres and found seroprevalence rates ranging by hospital from 0.8% to 31.2% (median = 3.6%). Higher rates were found in those hospitals situated where there was a high local area community cumulative incidence of COVID-19.[5] Apart from differences between countries or hospitals there are other factors that have been shown to influence rates of seropositivity which include gender,[5] ethnicity,[4, 5] and category of hospital work (e.g. housekeepers higher than intensive care staff).[4] In Belgium
in contrast a study found no relationship between direct clinical care of COVID-19 patients and seroprevalence rates, but instead found rates to correlate with reported household contacts.[23]

Comparison of seroprevalence rates between different studies is complicated by the varying performance of assays used to measure SARS-CoV-2 serum responses. There is currently no standardisation of assays and, as shown within the same seroprevalence study, the performance of seven different assay may differ widely.[19]

To reliably undertake trans-national comparison, this study centralised testing in a single laboratory. Staff in all participating centres were recruited irrespective of a history of clinical symptoms that could be construed as COVID-19 and thus should be considered as an unbiased sample of HCWs. All worked in paediatric facilities although some were embedded in larger adult facilities. No attempt was made to stratify between clinical facing and non-clinical staff as HCWs in paediatric facilities are generally not in high-risk environments as children represent only a small number (6%) of total COVID-19 admissions[24] and young children may be less likely to transmit virus than older individuals.[25] All cohorts in this study were dominated by females so gender differences in seroprevalence rates should not account for the differences noted in our cohorts and the mean ages and age ranges overlapped significantly and were thus comparable. The period for recruitment (both time taken in days and the months in which recruitment took place) differed between the cohorts although the majority of the recruitment took place between May and July either during or after the initial peak of disease in most countries. The UK site (London) enrolled two cohorts, one throughout May and one throughout June, both periods associated with similar numbers of new cases in London and relatively small differences in seroprevalence rates were seen between the two cohorts. This suggests that month of sampling is unlikely
to have had a major effect on the results of this study as most of the sampling in our eight participating centres occurred at a similar time in relation to the peak of disease. The time between symptom onset and blood testing for most of the cohorts were comparable, with the shortest mean time being 46 days in the South African cohort. As most assays detect antibody reliably at least 15 days after exposure,[26] most positives would have been captured in our cohorts although whether some staff may have lost IgG and thus sero-reverted is unclear. If this were the case though it would have affected the seven cohorts with a relatively longer time periods between symptoms and testing equally. As in other studies,[23] there was a discrepancy between proportion of staff reporting symptoms compatible with COVID-19 and the seroprevalence rate. This was most pronounced for the UK cohort during June 2020 where 76.6% reported compatible symptoms yet seroprevalence was “only” 16.93%.

Interestingly the seroprevalence rates for several of our HCW cohorts were similar to population-based estimates for the same city or region. The seroprevalence for the London cohort of HCWs (15.4-17%) was very similar to that measured for London based blood donors analysed by Public Health England in May and June (15-16%).[27] The Greek HCW cohort seroprevalence of 1.3% (but based on just one case), was similar to that found in a community based study of seroprevalence in residual sera in Athens (0.93%);[28] the absence of any IgG-positive HCWs in Austria similarly reflects a very low national seroprevalence of 0.15% ([http://www.statistik.at/web_en/statistics/PeopleSociety/health/123052.html](http://www.statistik.at/web_en/statistics/PeopleSociety/health/123052.html)), the absence of any IgG-positive HCWs in the Estonian staff reflects a very low seroprevalence in Tartu of 1.6%[29] while the low rate in the Lithuanian HCWs (0.66%) is below the 1.31% seen in a population based survey in Vilnius ([https://sam.lrv.lt/lt/nuoje/nuoje-pristatyti-])
Similarly, blood donor testing in Latvia has revealed a seroprevalence rate of 0.4% (Personal Communication). We have been able to locate one study of paediatric HCWs conducted in Spain and here too, seroprevalence was similar to the general population[10] and one study of physicians in a children’s hospital in Argentina were seroprevalence rates were 0.9%[11]. Together these data suggest that HCWs in paediatrics settings are no more likely than the general population to be seropositive for SARS-CoV-2 and household contacts may well be an important source of infection[23].

This study has a number of limitations. The cohort sizes differed significantly between centres although all but one enrolled >120 subjects. No attempt was made to adjust for sensitivity of the EDI assay as this study was comparative and the key outcomes was comparison of rates between the participating countries. The true seropositive rate might be slightly higher as the assay sensitivity locally was assessed as 92.4% with published sensitivity as low as 80%.[30] However all equivocal samples that were borderline in the EDI assay were re-assayed in the MSD assay that has very high sensitivity[14] increasing the overall sensitivity for detecting positives in these cohorts. As the cohorts separated into six countries with relatively low seropositivity rates and two with relatively high rates this limited the opportunity to correlate findings to factors measured by Google Mobility data or the individual government response to the pandemic.

Conclusion

This study shows that HCWs in paediatric facilities have seroprevalence rates that are in general similar to their local general population which in turn are closely related to the overall burden of COVID-19 in those areas where the hospitals are based. While this may be interpreted as the success of personal protection in paediatric healthcare facilities it is more likely to be the absence of nosocomial exposure or lack of transmission from infected
children to adults and thus a risk of exposure SARS-CoV-2 similar to the general public. The relatively small proportions of HCWs with antibodies in six of the eight participating centres suggests that as the current second wave of infections increases, staff working in paediatric facilities remain susceptible to infection. A recent outbreak amongst staff on a Paediatric Intensive Care Unit in Germany illustrates the serious consequences of infection for health services.[31] Measures to monitor and protect paediatric staff both at work and at home should be instituted and adhered to, to avoid shortages of critical staff in the health service while seronegative staff should be targeted for vaccination where available.

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Conflict of Interest Statement
None of the authors declare a conflict of interest

Contributions
DG conceived the study, OFP, II, VS, ET, MW, JL, HZ, LG and DZ all contributed to the design of the study and implementation in each site. LG, OFP, II, VS, ET, MW, HZ, LB, RI, JP, PJ, JL, ZFS, DI, LG and DZ were responsible for administering the study in each site. DG and MJ were responsible for the laboratory component. DG wrote the first draft and all authors contributed to subsequent versions.
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**Figure Legend:**

Seroprevalence estimates for SARS-CoV-2 anti-nucleocapsid IgG in healthcare worker cohorts in eight countries
<table>
<thead>
<tr>
<th>Country</th>
<th>No of samples</th>
<th>% Female</th>
<th>Age (mean, range)</th>
<th>Date of first nationally recorded COVID-19 infection</th>
<th>Sample collection date range</th>
<th>Number with Clinical Symptoms</th>
<th>Number with +VE PCR</th>
<th>Proportion with +VE PCR</th>
<th>Proportion of cohort with symptoms</th>
<th>If symptomatic, time between symptoms and blood test</th>
<th>Number +ve</th>
<th>Seroprevalence Rate (95% Confidence Intervals)</th>
<th>National COVID19 rate/100,000 at the time of sampling*</th>
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<tbody>
<tr>
<td>Austria</td>
<td>196</td>
<td>84.2</td>
<td>38, 22-65 yrs</td>
<td>25/02/2020</td>
<td>17-24/07/20</td>
<td>33</td>
<td>0</td>
<td>0</td>
<td>16.8</td>
<td>116 days</td>
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<td>0 (0-1.92)</td>
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<td>96.2</td>
<td>50, 19-71 yrs</td>
<td>27/02/2020</td>
<td>10-12/06/20</td>
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<td>0</td>
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<td>148.2</td>
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<td>19/06-16/07/20</td>
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<td>1.3 (0.23-7.0)</td>
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<td>19/05-20/06/20</td>
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<td>0</td>
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<td>28/02/2020</td>
<td>01-12/06/20</td>
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<td>2</td>
<td>0.67</td>
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<td>43, 21-65 yrs</td>
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<td>14/05-20/27/05/20</td>
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<td>10/06-17/08/20</td>
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<td>7.66</td>
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<td>772</td>
<td>15</td>
<td>0.86</td>
<td>44.0</td>
<td>64 days</td>
<td>269</td>
<td>15.34 (13.73-17.1)</td>
<td>390.1</td>
</tr>
<tr>
<td>United Kingdom</td>
<td>1134</td>
<td>72.9</td>
<td>37, 19-78 yrs</td>
<td>15/02/2020</td>
<td>01/06-30/06/2020</td>
<td>869</td>
<td>15</td>
<td>1.32</td>
<td>76.6</td>
<td>89 days</td>
<td>192</td>
<td>16.93 (14.86-19.22)</td>
<td>459.1</td>
</tr>
</tbody>
</table>
Table I: Overall demographics and results for the eight cohorts studied.

<table>
<thead>
<tr>
<th>Country</th>
<th>Average Google Mobility reduction in non-residential activity (%)</th>
<th>Oxford COVID-19 Government Response Tracker score (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Austria</td>
<td>-31</td>
<td>62</td>
</tr>
<tr>
<td>Estonia</td>
<td>-11</td>
<td>50</td>
</tr>
<tr>
<td>Greece</td>
<td>-37</td>
<td>63</td>
</tr>
<tr>
<td>Latvia</td>
<td>-19</td>
<td>70</td>
</tr>
<tr>
<td>Lithuania</td>
<td>-21</td>
<td>60</td>
</tr>
<tr>
<td>Romania</td>
<td>-42</td>
<td>50</td>
</tr>
<tr>
<td>South Africa</td>
<td>-40</td>
<td>90</td>
</tr>
<tr>
<td>United Kingdom</td>
<td>-33</td>
<td>75</td>
</tr>
</tbody>
</table>

Table II: Google Mobility and the Oxford COVID-19 Government Response Tracker for the eight participating countries
Figure 1: Seroprevalence estimates for SARS-CoV-2 anti-nucleocapsid IgG in healthcare worker cohorts in eight countries