Design and implementation of a national program to monitor the prevalence of SARS-CoV-2 IgG antibodies in England using self-testing: REACT-2 Study

ABSTRACT (180 of 180)

Data system The Real-time Assessment of Community Transmission-2 (REACT-2) study was funded by the Department of Health and Social Care to estimate community prevalence of SARS-CoV-2 IgG antibodies in England, UK.

Data Collection/Processing Random cross-sectional samples of adults were obtained from the National Health Service (NHS) patient list (near-universal coverage). Participants were sent a lateral flow immunoassay (LFIA) self-test, reporting the result online. Overall, 905,991 tests were performed (28.9% response) over six rounds of data collection (June 2020 - May 2021).

Data Analysis/ Dissemination Weighted estimates of LFIA test positivity (validated against neutralizing antibodies), adjusted for test performance, were produced at local, regional and national levels, and by age, sex, ethnic group and area-level deprivation. In each round, fieldwork occurred over two weeks with results reported to policymakers the following week. Results were disseminated as pre-prints and peer-reviewed journal publications.

Implications REACT-2 estimated the scale and variation in antibody prevalence over time. By using community self-testing and reporting, it produced rapid insights into the changing course of the epidemic and the impact of vaccine roll-out, with implications for future surveillance.

MAIN TEXT

The REal-time Assessment of Community Transmission-2 (REACT-2) Study sought to provide reliable and timely estimates of the prevalence of antibodies to severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) infection from random samples of the adult population of England.

DATA SYSTEM

This study involved 6 rounds of data collection, from June 20, 2020, to May 25, 2021 (Figure 1)

Name and sponsor

The REACT-2 study, funded by the Department of Health and Social Care in England and sponsored by Imperial College London.

Purpose (136)

The aim was to estimate the number and distribution of SARS-CoV-2 infections during the first and second waves of the COVID-19 epidemic in England by place and person, identify trends in antibody positivity, and subsequently measure the impact of vaccine roll-out on population antibody prevalence.

Public health significance

REACT-2 was established following the first wave of the COVID-19 epidemic in England when little was known about the extent of SARS-CoV-2 transmission in the community due to limited access to diagnostic testing outside of hospital settings. We provided estimates of cumulative community prevalence of SARS-CoV-2 IgG antibody test positivity with a rapid test and identified groups at highest risk of infection. In addition, we estimated the total number of individuals in England who had been infected, and the infection fatality ratio (IFR) overall and by age, sex and ethnic group. REACT-2 was designed to provide repeated snapshots of the

cumulative prevalence of test-positivity for antibodies, above the threshold of the rapid test, initially from infection and later from vaccination. These data fed directly into government to inform the public health response.

DATA COLLECTION/PROCESSING

In this study, random samples of adults in the community were invited to use at-home testing with a 'finger-prick' lateral flow immunoassay (LFIA) device, and to report the results plus demographic, behavioral and clinical details in an online or telephone survey.

A. Data sources and collection mode

Source population. Random cross-sectional samples of individuals aged 18-years and over in England were invited to participate. Our sample frame was individuals on the National Health Service (NHS) patient list, which includes name, address, age and sex of everyone registered with a general practitioner in England (almost the entire population).

Survey instruments. Data were collected through a web-based survey instrument designed and piloted with public input and hosted by our logistics partner, Ipsos. An invitation letter was sent by mail to named individuals who were directed to an online or telephone registration site where they could consent to the study. The registration form confirmed date of birth, and gathered additional information on household size and composition, occupation, education, and ethnic group (**Supplementary Material**). Eligible people (all except those with possible bleeding risk from use of a lancet) were directed to a consent form and asked for their email address and mobile (cell) phone number. Following registration, participants were sent a self-test LFIA kit, instruction booklet linked to an online video, and a link to a website (or phone

option) to complete a further user survey once they had completed the test. The survey instruments are available on the study website.

Finger prick antibody test. The LFIA (Fortress Diagnostics, Northern Ireland) was selected following evaluation of performance characteristics (sensitivity and specificity) against pre-defined criteria for detection of SARS-CoV-2 IgG (1,2). The LFIA uses the structural spike (S) protein of the virus as the target antigen for antibody-based detection. It was initially evaluated for (i) sensitivity in an NHS healthcare worker cohort known to have been infected with SARS-CoV-2 confirmed by RT-PCR, not hospitalized, at least 21 days earlier, and (ii) specificity using 500 pre-pandemic sera. Compared to results from at least one of two in-house ELISAs, sensitivity and specificity of finger-prick blood self-test were 84.4% (95% confidence interval [CI] 70.5%, 93.5%) and 98.6% (97.1%, 99.4%), respectively (1). The in-house ELISAs used were the spike protein ELISA (S-ELISA) and a hybrid spike protein receptor binding domain double antigen-bridging assay (hybrid DABA)(3). Further validation of the LFIA showed equivalent performance in a self-testing non-healthcare worker occupational cohort (4) and a healthcare worker and renal transplant patient cohort post-vaccination (5). We compared the self-test LFIA to a commercially available quantitative assay in 3758 participants, a majority of whom had been vaccinated or reported prior infection. The LFIA was less sensitive than the laboratory assay, being positive in 73.9% compared to 96.4% of participants; however in a subset of 250 samples, the LFIA correlated better with live virus neutralization (6).

Testing and reporting. The designs of the testing kit, instruction booklet and video were created by graphic designers specializing in healthcare, with input from 300 public volunteers in a pilot study which identified the need for improvements in elements of the kit, instructions and interpretation of results. This was followed by a larger pilot study of >14,000 randomly-selected members of the public which showed high levels of acceptability and usability (7). Using the instructions provided, participants carried out the LFIA using a finger-prick capillary blood sample, read the results, and reported them in the survey along with additional sociodemographic, behavioral and clinical details (**Supplementary Material**). Participants were asked to upload a photograph of the completed test.

B. Ethical procedures

Ethics. We obtained ethical approval from the NHS Health Research Authority South-Central Berkshire B Research Ethics Committee (IRAS ID: 28305). Participants gave individual consent to participate either online or by telephone. Approval for use of the test kit was obtained from the Medicines and Healthcare products Regulatory Agency (MHRA https://www.gov.uk/government/organisations/medicines-and-healthcare-products-regulatory-agency), with the caveat that the test was to be clearly labelled as for research purposes only, and that participants were given advice not to change their behavior in the light of the result.

Public involvement. A public advisory panel provided input into the design, conduct and dissemination of the study, and lay members sit on a data access committee governing further access to the data.

C. Population(s) and geographic coverage

Population. The target population was the adult population of England aged 18 years and over. We aimed to provide data at lower-tier local authority area (LTLA) level in England to aid local administrative and public health response to the epidemic. We included data for 316 of the 317 LTLAs in England (excluding Isles of Scilly), and by combining the two smallest with neighbouring areas we report on 315 areas. We also provided national and regional estimates of antibody positivity, and prevalence estimates for key demographic sub-groups including by age, ethnic group, socioeconomic status (as determined by an area-level deprivation score) and occupation. Estimates of weighted prevalence over the 6 rounds of the study are shown in **Figure 1**.

Sampling frame. The sampling frame was all adults 18 years and over who were registered with an NHS general practitioner in England. This information is held NHS England and provides near-complete coverage of the resident population.

Sampling strategy. Random samples were obtained from the NHS patient list, and individual invitations sent by post. The sample was stratified by LTLA with the aim of achieving similar numbers of participants in each local area. For round 6 (May 2021) we adjusted the sampling to achieve a boost of 70,000 people in age groups 55-64 and 65-74 years to include additional numbers post their first and second vaccinations, since vaccines were rolled out in order of decreasing age from December 2020 (8).

D. Unit of data collection and sample size

Unit of data collection. Data were collected at the individual level. The samples were non-overlapping until the final boosted round where some overlap with earlier rounds occurred, with 4950 people taking part twice over the six rounds.

Sample size and response rates. Over the six rounds of data collection from June 20, 2020 to May 25, 2021, a total of 905,991 completed tests were included from 3,134,353 invitations, giving an overall response rate (number of completed tests /number of invitations sent out) of 28.9%. The response rate varied by round (range: 26.3% to 33.5%), with completed tests ranging from 105,651 to 209,482 per round (**Figure 1**). The response rate also varied by sex, age, region and deprivation (Supplementary Table 1).

Sample size determination. In rounds 1 to 5 we aimed for 100,000 completed tests per round to provide meaningful information on the 315 LTLAs in England. The highest levels of uncertainty were in populations with low prevalence, where the point-antibody positivity could be so low that there were no positive tests in that area. With a total of 100,000 completed tests, we can exclude (95% confidence) a prevalence >1.7% in each LTLA recording zero positive tests. In Round 6 we aimed for a total sample size of 240,000 test results including, as noted, a boost of 70,000 people in age groups 55-64 and 65-74 years powered to detect a clinically important difference in outcome (relative risk 0.5 for hospitalization) between test-positive and test-negative individuals.

Completeness. By design, we aimed for approximately equal numbers of participants in the 315 LTLAs in England. The achieved samples at LTLA level ranged from 200 to 598 in rounds 1 to 5, and 517 to 802 in round 6 with the boosted sample. We achieved sufficient data by round to estimate prevalence by age, region and other key demographic groups including ethnic group, deprivation index and occupation.

Generalizability. Our study had lower response among men, youngest and oldest groups, people from minority ethnic groups and in more deprived areas (Supplementary Table 1). Unequal participation is observed in almost all population surveys. To account for differential response, we weighted the data at each round to be representative of England as a whole, although this may not fully correct estimates.

E. Surveillance design and frequency of data collection

Surveillance design. This was a serial cross-sectional design, randomly-selected, with largely non-overlapping samples across six rounds of the study. The key was use of at-home self-testing and reporting of the results from a point-of-care rapid test, which enabled results to be obtained at scale and disseminated quickly. Most data collected were reported by participants, including history of COVID-19, comorbidities, and vaccination. However, where we had specific consent for data linkage, we were able to link to routine health data to confirm vaccination status and obtain outcome data (hospitalizations, deaths).

Frequency of data collection. The study was initially commissioned to estimate the total number of people who had been infected with SARS-CoV-2 in the first wave in England which peaked in March 2020 and declined rapidly following the introduction of a strict lockdown on 23 March

(9). The first round took place at the end of June 2020, followed by three more rounds (2 – 4) at 6-weekly intervals in July/August, September and October 2020 (**Figure 1**). There was a 2-week reporting window for participants to upload their results, and the overwhelming majority performed the test and reported the results in the first few days of those periods. The final two rounds took place after a gap of 3 and 4 months (January and May 2021). The rounds were timed to capture the prevalence and trends in population antibody positivity (i) following the first wave (rounds 1 and 2), (ii) during the emergence of the second wave (rounds 3 and 4), and (iii) to assess the impact of vaccination (rounds 5 and 6). No further rounds were commissioned.

Key data elements and data quality/editing

Prevalence estimates. Prevalence was calculated as the proportion of individuals with a positive IgG test result on the LFIA, adjusted for test performance using:

$$p = (q + specificity - 1) / (sensitivity + specificity - 1)$$

where p = adjusted proportion positive, q = observed proportion positive (10).

Prevalence estimates (and 95% confidence intervals) were weighted to account for the geographic sample design and for variation in response rates to be representative of the population (18+ years) of England (Supplementary Table 1). The approach used random iterative method (RIM) weighting (11) to adjust to population estimates for age; sex; Index of Multiple Deprivation (IMD) decile (12); LTLA; ethnic group. The weighting approach is based on that described in Elliott et al (13), but for 7 rather than 9 age categories.

We used logistic regression to identify sociodemographic variation in antibody positivity by estimating the odds ratio (OR). An OR >1 indicated that the group was more likely to have higher prevalence of antibody test-positivity relative to the reference group per sociodemographic variable. Models were adjusted for age, sex and region, and additionally for ethnic group, deprivation, household size and occupation.

We estimated the Infection Fatality Ratio (IFR) from the total number of COVID-19 deaths among adults in England (14) divided by our estimate of the total number of SARS-CoV-2 infections since the start of the epidemic until mid-July 2020. This was estimated by multiplying the weighted and adjusted antibody prevalence by the mid-year population size at ages 18+ years in England. We obtained an overall IFR estimate of 0.90% (0.86, 0.94) as well as estimates stratified by age, sex and ethnic group (15).

LFIA self-testing procedure.

The LFIA requires a blood sample from a finger-prick and produces a test result after 10 to 15 minutes. The test kits sent to participants included 1 LFIA device, 1 bottle of buffer solution, 2 pressure-activated 23G lancets, 1 alcohol wipe and a 1 mL plastic pipette, alongside an instruction booklet with weblink to a video.

The key visual features of the Fortress SARS-CoV-2 LFIA device include the test result window and blood sample well (Figure 2). The result window has an initially blue control line, which will remain if the test is unsuccessful (Invalid). In a successful test, the control line turns red, and if

IgG antibodies are present in the blood sample above a threshold, a secondary line will appear below the control. There is also a line indicating IgM, but this performed poorly in our initial laboratory evaluation, and was not analysed. Participants were informed that results were not reliable at an individual level.

Data security. Data were transferred securely from Ipsos to Imperial College London and held on secure servers in an ISO27001 environment managed by the School of Public Health. Study participants were assigned a study ID and data stripped of identifying information for the statistical analyses; only a small number of named and designated individuals have access to identifying information in line with a published privacy policy[1] and compliant with the UK Data Protection Act 2018, which is the UK's implementation of the General Data Protection Regulation (GDPR https://www.gov.uk/data-protection).

Managing disclosure risks. To protect confidentiality, individual data are not released, and tabular data are suppressed if there are fewer than 5 entries in a cell where one or more person is a positive for SARS-CoV-2 IgG on LFIA.

DATA ANALYSIS/DISSEMINATION

[1] Privacy notice Imperial College London: https://www.imperial.ac.uk/media/imperial-college/institute-of-global-health-innovation/REACT_1_Round_19_Antigen_Privacy_Notice_PUBLIC.pdf and DHSC: https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/800049/Privacy_Notice_v2_.pdf

The results of REACT-2 per round were fed weekly into government to provide situational awareness and inform public health policy. In addition, we placed REACT-2 data and results into the public domain in near real-time (through preprints and media press-releases), thus informing both the public and the international scientific community of emerging data on prevalence of SARS-CoV-2 antibody test-positivity.

A. Interpretation issues

During the study period, we observed a gradual fall in response rates, from a high of 33.5% in round 1 (June, 2020) carried out following the first wave in England, to 26.3% in round 5 (January, 2021) conducted in the early stages of vaccine roll out. In round 6, the response rate rose to 28.0%, reflecting the boosted sample of individuals aged 55-74 years who generally had high response rates to our surveys. Our surveys also had lower response among people from minority ethnic groups and in more deprived areas. We re-weighted the sample in each round to account for differential variation in response to be representative of the population (18+ years) of England as a whole, although this may not have overcome unknown participation biases.

We used a qualitative ('Yes/No') at-home self-administered LFIA on a finger-prick capillary blood sample instead of more resource-intensive "gold standard" quantitative laboratory tests performed on venous blood samples. To demonstrate the validity of this approach we conducted extensive evaluation of the selected LFIA which showed it to have acceptable performance (sensitivity and specificity) in comparison with confirmatory laboratory tests (1). We took steps to measure and improve usability, including ability to perform and read an LFIA

test at home (4,7). By adjusting our survey results for known LFIA performance, we demonstrated that, despite not meeting regulatory standards for clinical use in individuals, self-testing and reporting using LFIAs provide a valid tool for obtaining reliable community-wide prevalence estimates in a cost-effective manner, rapidly and at scale.

For those with a self-reported clinical history of confirmed or suspected COVID-19, there was potential for reporting bias as respondents were not blinded to their test results; however, there was high concordance of self-test with clinician-read results. To support ongoing quality assurance for the self-tests, we designed an Automated Lateral Flow Analysis (ALFA) computerised pipeline using machine learning, computer vision techniques and signal processing algorithms to analyse the uploaded images of the test (16), finding high concordance with self-test results.

Our study demonstrated a substantial decline (26.5%) in population antibody test-positivity over three months between rounds 1-3 (June 20th to September 28th, 2020), indicating antibody waning 3 and 6 months after the first wave of infections (Figure 2)(17). To exclude the possibility this could be due to differences in LFIA batch, we compared the laboratory performance of the LFIAs used in rounds 1 and 2 (where we had seen the strongest decline in positive tests) and found no difference between the two rounds.

Linkage ability

Data linkage (based on unique NHS number) to vaccination status (vaccine type and date) and outcome data (hospitalizations, deaths) is available for participants who consented to linkage to their health records.

Data release/accessibility

Access to REACT-2 individual-level data is restricted to protect participants' anonymity. Summary statistics, descriptive tables and code from REACT-2 are available on Github, and study materials for each round are on the study website https://www.imperial.ac.uk/medicine/research-and-impact/groups/react-study/for-researchers/react-2-study-materials/.

Key references/other information

We published our initial protocol (18) and our key findings during the 11 months of fieldwork (15,17,19–21), including clinical and laboratory evaluation of antibody tests and feasibility studies of at-home self-testing and reporting using LFIAs, (2,5–7,16), in preprints and peer-reviewed journal publications. Links to all our publications are given on the study website https://www.imperial.ac.uk/medicine/research-and-impact/groups/react-study/real-time-assessment-of-community-transmission-findings/ and included for reference in the **Supplementary Material**.

IMPLICATIONS

REACT-2 provided reliable and robust estimates of population prevalence of SARS-CoV-2 IgG antibody test-positivity during the first two waves of the epidemic of COVID-19 and the initial stages of vaccine roll out in England. It demonstrated high feasibility and acceptability of using

295 at-home self-administered LFIA tests (self-reported and uploaded photo for verification) as a
296 means of providing reliable, cost-effective community-wide prevalence estimates rapidly and at
297 scale. This contrasts with the use of quantitative laboratory assays which require blood to be
298 collected, transported and processed in a laboratory.

299 REACT-2 confirmed early reports that SARS-CoV-2 disproportionately affected people from
300 disadvantaged and minority ethnic groups in England, as well as health and care workers
301 (**Figure 4**), suggesting that the higher hospitalization and mortality from COVID-19 in these
302 population groups reflected higher rates of infection. We found no difference in estimated IFR
303 between people of broad ethnic categories (Black, Asian, white) when stratified by age and sex
304 (15). Based on participant responses to questions about onset of prior COVID-19 symptoms, we
305 were able to reconstruct an epidemic curve for infection in early 2020 which closely matched
306 but slightly pre-dated the curves of hospitalizations and deaths (15). This gives context validity,
307 and provides an indication of the size and shape of the first and second waves (Figure 3). The
308 epidemic curve was replicated in each round providing further validation of the approach
309 (15,17,19,20).

310 We also provided timely information on changes in the prevalence of antibody positivity over
311 time due both to natural infection and vaccination (**Figure 1**). The observed decline in
312 population antibody positivity following the first wave (**Figure 4**) supported emerging data on
313 SARS-CoV-2 that indicated a decrease over time in antibody levels ('waning') in a proportion of
314 individuals followed in longitudinal studies(22). Prior to vaccination, we observed waning of
315 26.5% over three months, with the biggest decline in older people(17). In the later rounds, by

316 tracking antibody test-positivity to COVID-19 following vaccination and showing differential
317 waning, our study provided key data underpinning vaccination policy and contributed to
318 recommendations regarding groups who might benefit from additional vaccine doses (20,21).

319 Finally, the success of REACT-2 was underpinned by rapid public involvement at every stage.
320 Public volunteers and a diverse advisory panel provided input into the design and conduct of
321 the study. Their desire to support the national response shows that public involvement is both
322 possible and necessary during periods of emergency response.
323
324 Antibody self-testing at home is feasible, acceptable and can provide essential data to policy
325 makers within days. In order to roll this out in a timely fashion in future pandemics, it is
326 important to invest in the necessary technologies and infrastructure (23) including capacity for
327 test production, putting in place logistics of implementation, together with capability in study
328 design and data analysis.

Contributors

[Lead authors] drafted the article. The other authors critically reviewed the article and provided comments. All authors agreed to submission for publication.

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Note. The funders had no role in the design and conduct of the study; collection, management, analysis, and interpretation of the data; and preparation, review, or approval of this article.

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Figure legends

Figure 1. REACT-2 study timeline from June 20, 2020 to May 25, 2021 over 6 rounds of data collection. For each round we report the number of invitations sent (I), the number of participants registered (R), the number of completed self-tests reported (T), response rate (RR) as defined by the number of self-reported completed LFIA tests over the number of invitations sent, completion rate (CR) as defined by the number of completed tests over the number of kits sent out, and the prevalence of antibody positivity, adjusted for test characteristics and weighted to the adult population of England. Note the reported response rates are conservative as i) not all invitations would have been received (or opened) by the potential participants, and ii) recruitment was stopped once the required sample size had been reached.

Figure 2. Diagram of lateral flow immunoassay (LFIA) kit with guide to reading and reporting the result The test cassette is shown on the left indicating the buffer solution and blood sample wells, and the test result window. The detail of the test result window is on the right indicating what invalid, negative and positive results look like. The wording from the questionnaire on how to report the result is reproduced below the figure.

Figure 3. Reconstruction of COVID-19 epidemic curve by week of symptom onset reported by REACT-2 participants (top) alongside national data on admissions and deaths from COVID-19 in England (bottom). The top chart shows two curves: the solid line includes date of onset for all cases of COVID-19 reported by participants; the dashed line is limited to those who had a positive lateral flow immunoassay (LFIA) test result in the REACT-2 study.

436

437 **Figure 4**. Antibody prevalence with confidence intervals by round for rounds 1 to 4 (pre-

438 vaccination), in the sample overall, and stratified by sex, age, ethnic group and employment.

439 Estimates are adjusted and weighted except for employment where data are not available for

440 weighting.

Figure 1 REACT-2 study timeline with number of invitations, registrations, tests completed, response rate, completion rate and weighted prevalence, June 2020 to May 2021

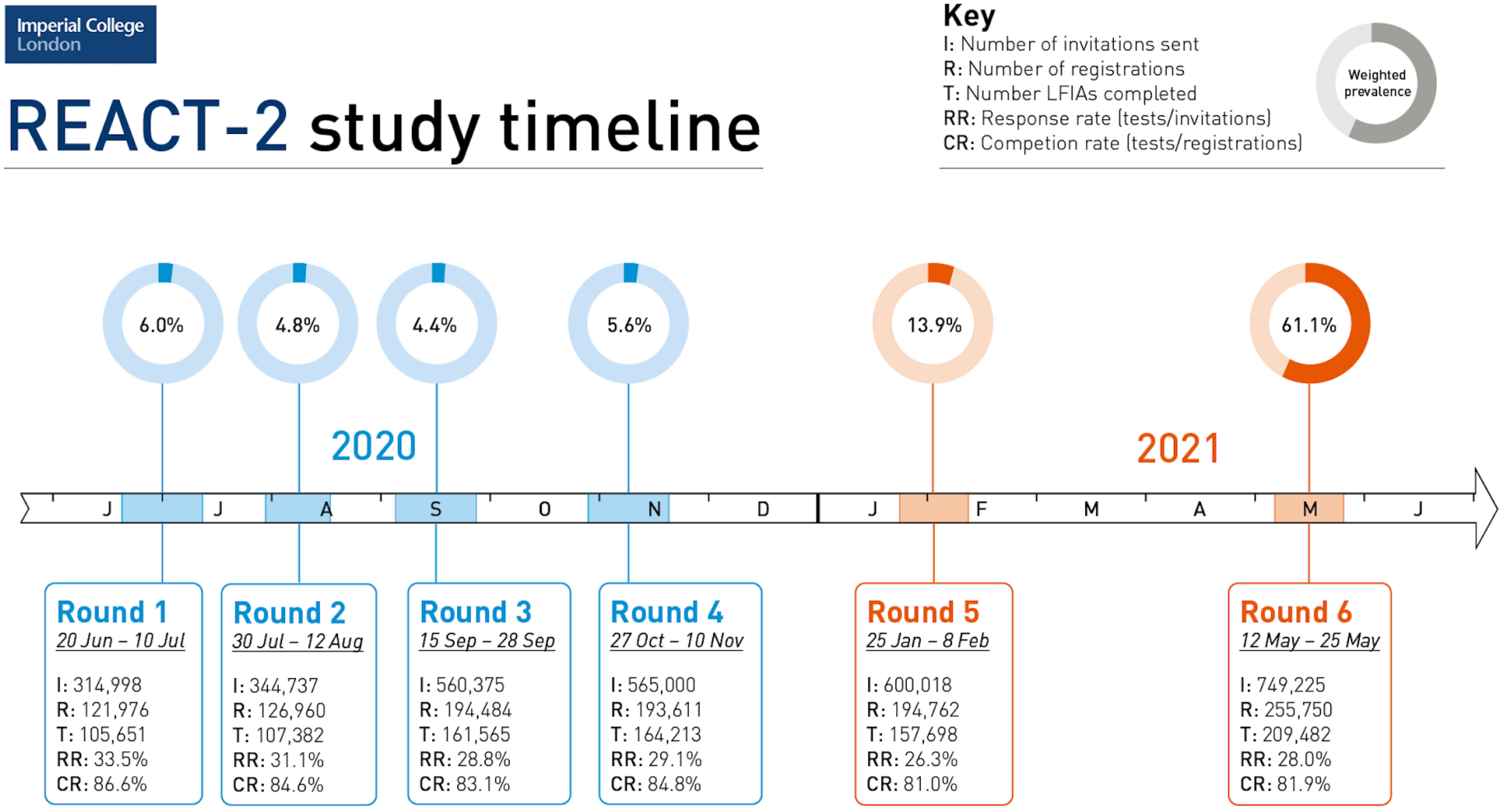
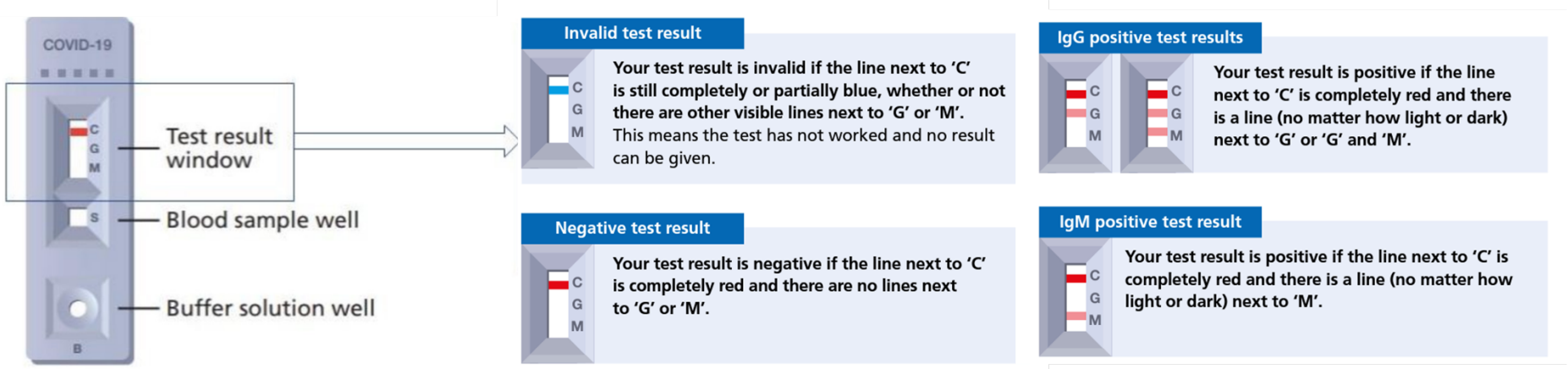


Figure 2: Diagram of lateral flow immunoassay (LFIA) kit with guide to reading and reporting the result



Wording from the questionnaire:

Step 9 of the instruction booklet shows different test outcomes. Based only on the photo you took and what the test looked like after 10-15 minutes, which number corresponds to your test result?

Note: How light or dark the colour of the line is next to G and/or M will vary. Therefore, any shade of colour next to G and/or M should be reported if the line next to C is red.

O (Negative)	Red line next to C only. No lines next to G or M.
1 (Ig M Positive)	Red line next to C and red line (no matter how light or dark) next to M. No line next to G.
2 (Ig G Positive)	Red line next to C and red line (no matter how light or dark) next to G. No line next to M.
3 (Ig G Positive)	Red line next to C and red lines (no matter how light or dark) next to G and M.
4 (Invalid)	Line next to C is completely or partially Blue. This means the test is invalid even if there are red lines next to G or M.
5	Can't tell what the result is

Figure 3 Reconstructed epidemic curve from REACT-2 participants reporting date of COVID-19 symptom onset, alongside national hospitalization and death data

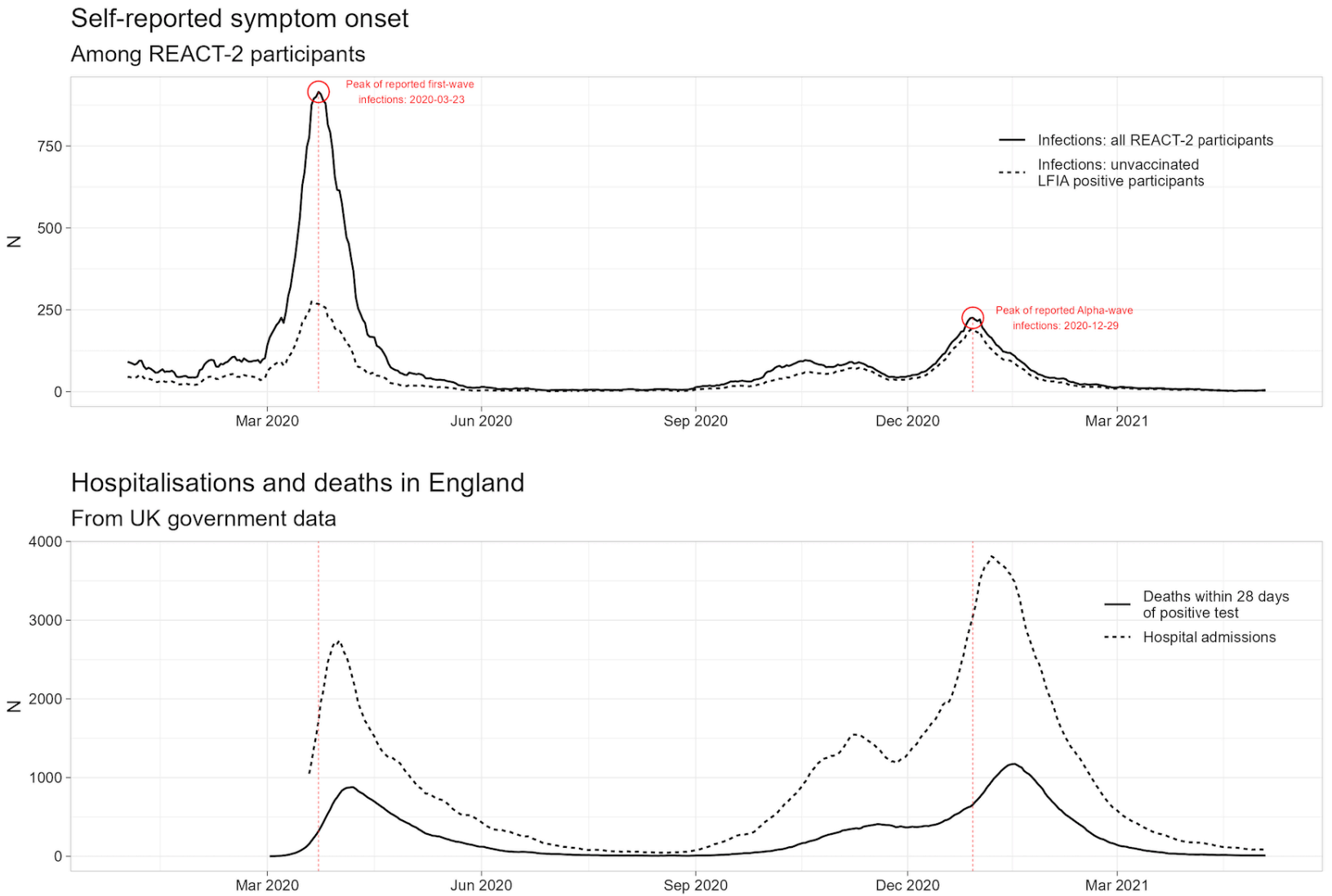


Figure 4: Antibody prevalence by round overall and by sex, age, ethnic group and employment for rounds 1 to 4 (pre-vaccination)