Supplementary appendix

This appendix formed part of the original submission and has been peer reviewed. We post it as supplied by the authors.

This online publication has been corrected. The corrected version first appeared at thelancet.com on Nov 25, 2020.

Supplement to: The National SARS-CoV-2 Serology Assay Evaluation Group. Performance characteristics of five immunoassays for SARS-CoV-2: a head-to-head benchmark comparison. Lancet Infect Dis 2020; published online Sept 23. https://doi.org/10.1016/S1473-3099(20)30634-4.
Supplementary material

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Supplementary Methods

A. Systematic review

We considered any article published in English, German, French, Spanish or Italian. Screening was performed
by a single reviewer, using a screening template to exclude studies if they were not focused on determining
SARS-CoV-2 serology, did not evaluate sensitivity/specificity for (an) assay(s), or did not evaluate assays
included in our head-to-head; replicate references across databases were de-duplicated. Data extraction for
relevant studies was completed by two extractors using a template table (in line with PICOS [Participants,
Interventions, Comparisons, Outcomes, Study design]) which included publication details, a description of the
cohort samples investigated, the sample sizes used to generate metrics, sensitivity and specificity for the
relevant assay being evaluated, and additional freetext notes (see Supplementary Methods/Supplementary
dataset SD1/PRISMA checklist).

The expanded search terms in PubMed were as follows:

("severe acute respiratory syndrome coronavirus 2"[Supplementary Concept] OR "severe acute respiratory
syndrome coronavirus 2"[All Fields]) OR "sars cov 2"[All Fields]) AND ((((((((("elisa s"[All Fields] OR
For the PubMed search, the workflow is shown below:

For the BioRxiv/MedRxiv searches, the workflow is shown below:

Article screening results can be found in Supplementary Dataset SD1.
B. Cohorts from which samples were obtained

‘Known negative’ samples
- Oxford BioBank, Oxfordshire, UK; Oxfordshire Clinical Research Ethics Committee 08/H0606/107+5

‘Known positives’ samples
- Gastro-intestinal illness in Oxford: COVID substudy [Sheffield REC, reference: 16/YH/0247]
- ISARIC/WHO Clinical Characterisation Protocol for Severe Emerging Infections [Oxford REC C, reference 13/SC/0149]
- Sepsis Immunomics project [Oxford REC C, reference:19/SC/0296]
- Volunteer plasma donors being screened for convalescent plasma studies by NHS Blood and Transplant (NHSBT; RECOVERY [Cambridge East REC (ref: 20/EE/0101]) and REMAP-CAP [EudraCT 2015-002340-14] studies).

C. Oxford immunoassay (OIA) validation and calibration

Schematic of OIA and workflow

Plate layout - schematic

|   | 1   | 2   | 3   | 4   | 5   | 6   | 7   | 8   | 9   | 10  | 11  | 12  | 13  | 14  | 15  | 16  | 17  | 18  | 19  | 20  | 21  | 22  | 23  | 24  |
|---|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
|   | ![Sample Plate Schematic](image)

Samples
- 1:400 dilution
- NHSBT high plasma standard (donor ID: 10062)
- NHSBT medium plasma standard (donor ID: 10065)
- NHSBT low plasma standard (donor ID: 10063)
- Neophiles (pre-pandemic) control
- NHSBS reagent (not always added)
Assay modifications
The assay was implemented as in (1), but with the following minor modifications:

- A 4-times instead of 5-times wash after incubating the sera
- Incubating with the secondary antibody for 90mins instead of 60 mins
- Adding a further 2-times wash with PBS after the secondary antibody had been washed away three times with PBS-T
- Incubating with 20μL of QuantaRed™ Enhanced Chemifluorescent HRP Substrate Kit (Thermo Scientific, Waltham Massachusetts, USA) for three instead of four minutes before the addition of 2μL of the stop solution

Calibration of the OIA assay
A total 8 milk ‘blank’ wells were included in each plate to determine a background reading which was subtracted from the raw signal per well to obtain a net reading.

A panel of dilution series made from the 3 NHSBT controls and the monoclonal antibody (CR3022), run on each 384 well plate, were used to calibrate readings between plates and batches. NHSBT 10061 was used at 1:25, 1:50, 1:100, 1:200, 1:400, 1:800, 1:1600, 1:3200, NHSBT 10062 at 1:25, 1:400, 1:800, 1:1600, 1:3200 and NHSBT at 1:25, 1:50, 1:100, 1:200, 1:400, 1:800, 1:1600 dilutions. CR3022 was used at concentrations of 3, 1, 0.3, 0.1, 0.03, 0.01, 0.003, 0.001 µg/ml. The majority of controls were included in duplicate (n=50 overall). An additional 5 NIBSC controls were run on a subset of runs. A single negative control, BD01 (a pre-pandemic blood donor serum provided by NHSBT) was also included on all plates.

A reference set of assay values for these controls were determined from data obtained on 3rd June 2020. All runs were converted into “3rd June” units using a natural cubic spline based linear regression model. For visualisation, values obtained for each control dilution within each 384 well plate were plotted on the x-axis and the reference values plotted on the y-axis. A linear regression model was fitted between the two, transforming the values on the x-axis using a 3-knot natural cubic spline (see Figure below). Model parameters were then used to convert net readings for all samples in the plate to normalised net readings.

Example mapping between net readings and normalised reference readings for control sample dilutions.

After initial model fitting a heuristic check for outliers was performed searching for differences between the normalised net readings and reference values >800,000 units. Such points were excluded, typically ≤2 out of a total of 50 controls, before re-fitting a final model.

Derivation of OIA diagnostic thresholds
We derived thresholds for the Oxford immunoassay (OIA) using an independent set of derivation samples:

- Known positives; defined by SARS-CoV-2 RT-PCR positive nose/throat swab; n=120
  - Acute; n=21
    - >10-28 days from symptom-onset; n=21
  - Convalescent; n=99 individuals
- >28 days from symptom-onset (from Oxfordshire); n=18
- >28 days from PCR-positive result (obtained from NHSBT); n=81 samples
- MERS-CoV anti-sera; n=1
- Other respiratory virus infections; n=23
  - Acute seasonal coronavirus-positive samples (≤28 days post-respiratory PCR [BioFire FilmArray RP panel]); n=6
  - Convalescent seasonal coronavirus-positive samples (>28 days post-respiratory PCR [BioFire FilmArray RP panel]); n=11
  - Acute non-coronavirus respiratory virus infections; n=6 (≤28 days post-respiratory PCR [BioFire FilmArray RP panel])
- Pre-pandemic controls n=1205
  - Serum samples taken from in- or outpatients having health monitoring, or samples as part of clinical management; n=954
  - Pre-pandemic blood donors recruited by NHSBT; n=251

We used a prespecified specificity target of 99% to set a threshold for determining a threshold for presence of antibody. The distribution of results obtained is shown in the figure below.

**OIA diagnostic threshold derivation samples**

At ≥10 days post symptom onset, excluding the MERS anti-sera (which was positive) but including the 23 samples from individuals with other viral infections, derivation sensitivity and specificity (at a normalised threshold of 8 million units) were 100% (120/120; 95% CI: 97.0-100.0%) and 99.6 (1223/1228; 95% CI: 99.1-99.9%) respectively.

**Other methods**

* Sensitivity was calculated using the following equation:
  \[
  \text{Sensitivity} = \frac{\text{Number of true positive tests}}{\text{Number of true positive tests} + \text{Number of false negative tests}}
  \]

* Specificity was calculated using the following equation:
  \[
  \text{Specificity} = \frac{\text{Number of true negative tests}}{\text{Number of true negative tests} + \text{Number of false positive tests}}
  \]

* The positive predictive value (PPV) was calculated from the following equation:
  \[
  \text{PPV} = \frac{\text{Sensitivity} \times \text{prevalence}}{\text{Sensitivity} \times \text{prevalence} + (1 - \text{specificity}) \times (1 - \text{prevalence})}
  \]
The negative predictive value (NPV) was calculated from the following equation:

\[ \text{Specificity} \times (1-\text{prevalence}) / [(1-\text{sensitivity}) \times \text{prevalence} + (\text{specificity} \times (1-\text{prevalence}))] \]
**Supplementary Tables**

Table S1. Summary of serum/plasma samples used for head-to-head analysis of five immunoassays for the detection of SARS-CoV-2 antibodies. This reflects the details for samples that were evaluated across all assays.

| Group          | Source                                                                 | Number of samples | Days from symptom onset, median (IQR; min, max; number of samples) | Days from PCR-positive test, median (IQR; min, max; number of samples) |
|----------------|------------------------------------------------------------------------|-------------------|---------------------------------------------------------------------|---------------------------------------------------------------------|
| Known negative | Healthy individuals 30-50 years of age, collected between 2015-2018 in Oxfordshire (Oxford BioBank, [www.oxfordbiobank.org.uk](http://www.oxfordbiobank.org.uk)) | 976               | n/a                                                                 | n/a                                                                 |
| Known positive | Healthcare workers and patients ≥18 years of age at Oxford University Hospital NHS Foundation Trust, Oxfordshire, UK | 158               | 36·5 (28-53; 20, 73; n=158)                                          | 27 (4-58; 3, 59; n=105)                                             |
| Known positive | Volunteer plasma donors ≥18 years of age via NHS Blood and Transplant (NHSBT), across the UK | 378               | All samples ≥28 days post-symptom onset*                               | 44 (40-49; 32, 82; n=378)                                           |

*Although specific data on time from symptoms is not available for this group, all donors had to have been at least 28 days post-symptom onset to be eligible for sampling: see [https://www.nhsbt.nhs.uk/plasma-trial/](https://www.nhsbt.nhs.uk/plasma-trial/)*
**Table S2. Summary of the commercial immunoassays evaluated.** Information presented is based on the product literature released by each manufacturer, using versions active on 8-June-2020 when our protocol was finalised.

| Assay and analyser used | Viral target and antibody type | Sample type | Sensitivity (95% CI) on samples taken ≥14 days post-symptom onset/post-positive RT-PCR, number of samples | Specificity (95% CI), number of samples | Manufacturers’ thresholds |
|-------------------------|--------------------------------|-------------|-------------------------------------------------------------------------------------------------|-----------------------------------------|---------------------------|
| Abbott SARS-CoV-2 Immunoassay, Architect i2000SR | Nucleocapsid protein, IgG | Serum, serum separator tube and plasma (ACD, CPD, CPDA-1, dipotassium EDTA, tripotassium EDTA, lithium heparin, lithium heparin separator tube, sodium citrate, sodium heparin) | 96.77% (90.86, 99.33), 88 (≥14 days post-symptom onset) | 99.63% (99.05, 99.90), 1070 | Negative: <1.4 Positive: ≥1.4 |
| DiaSorin LIAISON® SARS-CoV-2 S1/S2 IgG, LIAISON® XL | Spike protein S1/S2, IgG | Serum, plasma (sodium heparin, lithium heparin, potassium EDTA) | 97.56% (87.40, 99.57), 14 (≥15 days from diagnosis (RT-PCR)) | 98.5% (97.6, 99.2), 1090 | Negative: <12.0 AU/mL Equivocal: 12.0 ≤ x <15.0 AU/mL Positive: ≥15.0 AU/mL |
| Roche Elecsys® Anti-SARS-CoV-2, Cobas e 411 | Nucleocapsid protein, Total antibody | Serum collected using standard sampling tubes, Li-heparin, K2-EDTA and K3-EDTA plasma | 100% (88.1, 100), 29 (≥14 days from diagnosis (RT-PCR)) | 99.81% (99.65, 99.91), 5272 | Non-reactive: <1.0 Reactive: ≥1.0 |
| Siemens SARS-CoV-2 Total (COV2T), Atellica Solution immunoassay analyzer | Spike protein S1 RBD, Total antibody | Serum and plasma (potassium EDTA and lithium heparin) | 100.00% (91.59, 100.00), 42 (≥14 days from diagnosis (RT-PCR)) | 99.82 (99.34, 99.98), 1091 | Non-reactive: <1.0 Reactive: ≥1.0 |
| Publication DOI/URL, first author, date | Cohort | Sample number | Reactive/ positive | Specificity % (95% CI) | Notes |
|----------------------------------------|--------|---------------|-------------------|------------------------|-------|
| Abbott  
10.1128/JCM. 01029-20, Brecher S et al, 27/May/2020 | RT-PCR-negative for SARS-CoV-2, PCR (Respiratory Panel 2, Film Array, BioFire Diagnostics) positive for other seasonal coronaviruses. Plasma samples taken >4 weeks after the positive respiratory PCR | 9 | 0 | 100% (66.3,100) | Very small study focused on analytical specificity in the context of SARS-CoV-negative (by RT-PCR) individuals. Elderly male population as the study was undertaken in three regional Veterans Affairs (VA) institutions in the US. |
| 10.1093/clinchem/hvaa120, Tang MS et al, 18/Jun/2020 (as BioRxiv preprint on 10/May/2020) | Control specimens included: 80 patients symptomatic but PCR negative for SARS-CoV-2; 50 serum specimens collected and frozen in 2015 before the emergence of SARS-CoV-2; 5 specimens from patients with other coronaviruses confirmed by molecular testing but PCR negative for COVID-19 (including Coronaviruses HKU1, NL63, and 229E); 4 specimens from patients with Influenza A or B. 14 specimens with potentially interfering antibodies were also included: 5 were positive for CMV IgG, 3 were positive for EBV VCA IgG, 3 were positive for EBV VCA IgM, 2 were positive for both EBV VCA IgG and IgM, and 1 was positive for rheumatoid factor | 153 | 1 | 99.4% (96.41, 99.98) | The single false-positive was a patient with a consistent syndrome and "prolonged exposure to a family member with PCR confirmed COVID-19" i.e. a likely Covid-19 case that was falsely negative by RT-PCR, highlighting the difficulty with using post-pandemic samples as a "known negative" group for evaluating specificity. |
| 10.1128/JCM. 00941-20, Bryan A et al, 7/May/2020 | Pre-pandemic specimens 2018-2019 sent to the clinical laboratory | 1020 | 1 | 99.9% (99.5, 100) | Proposed the use of AUC analysis to adjust thresholds: “These analyses indicated that optimal thresholds for the serologic diagnosis of SARS-CoV-2 was 1.42-1.49 at ≥ 17 days from symptom onset (sensitivity and specificity 100%); 0.7 at ≥ 14 days from onset (Sens 97.9%, Spec 99.6%); 0.7 at ≥ 10 days from onset (Sens 94.4%, Spec 99.6%); and 0.7 at ≥ 7 days from onset (Sens 88.0%, Spec 99.6%)” Pre-pandemic negative group represented samples submitted from 1010 individuals for HSV Western blot serology evaluation - i.e. likely to represent a biased subset of the population in whom HSV testing was being performed. |
### DiaSorin Liaison SARS-CoV-2 S1/S2 IgG

| Total | Confounders: COVID-19-negative but who had other viral, bacterial, parasitic or autoimmune pathologies | Other coronavirus infection: COVID-19-negative patients but positive to another strain of coronavirus (NL63 strain and OC43 strain) | Healthy subjects |
|-------|-------------------------------------------------|-----------------------------------|-----------------|
| 81    | 73                                              | 2                                 | 6               |

**Proposed the use of ROC analysis to improve thresholds resulting in specificity of 99% (95% CI: 93%–100%) and sensitivity of 100% (95% CI: 92%–100%).**

No breakdown by subgroup.

### Pre-pandemic serum samples (routine laboratory, 2011)

| Individuals with other coronavirus infections | SARS-CoV-2 RT-PCR negative patients | SARS-CoV-2 microneutralisation antibody negative samples | All negatives |
|-----------------------------------------------|------------------------------------|------------------------------------------------------|--------------|
| 1140                                          | 10                                 | 50                                                  | 1380         |

**98.5% (97.6, 99.1)**

No breakdown by subgroup.

### Pre-pandemic samples from 2018-2019

| Individuals with other infections/autoantibodies including: | 70 | NA | 94.9% (NA) |
|------------------------------------------------------------|----|----|------------|
| 39 samples had autoantibodies                               |    |    |            |
| 3 acute EBV                                                 |    |    |            |
| 4 acute coronavirus                                          |    |    |            |
| 35 from patients with respiratory infection                 |    |    |            |

**Small sample size, with all samples coming from individuals with other infections/autoantibodies, confidence intervals not estimated.**

Unclear which subset of 70 samples were used to evaluate the DiaSorin assay (insufficient sample to test all samples across all immunoassays included in the evaluation).

### Total 191 individuals (negative by 3 sequential RT-PCR results)

| n=7 negative | NA | 96.8 (89.0-99.6) |

**Number of samples and individuals tested not**
| 0.05.22.20106 | 328, Plebani M et al, 26/May/2020 |
| Blood donors from 2015 | Patients with autoimmune disease | Healthcare workers |
| samples | 131 (pos and neg) samples from 271 individuals | 101 individuals |
| 19 individuals | 71 individuals |
| clear. Not specified which subset of positive/negative individuals/samples used. | Using optimised thresholds: 88.9% (78.4-95.4) |

| 05.2020 | GeurtsvanKessel CH et al, 05/May/2020 |
| Serum and plasma from people exposed to a human coronavirus (HCoV-229E, NL63 or OC43), SARS, MERS, or with a range of other respiratory viruses, and patients with recent CMV, EBV or M. pneumoniae infection. (69/147 negatives run on Liaison) |
| 69 samples |
| 1 |
| 98.55% (92.24, 99.93) |
| Evaluation on samples from patients with other infections only. |

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| https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/887222/PHE_Evaluation_of_Roche_Elecsys_anti_SARS_CoV_2.pdf, PHE, 18/May/2020 |
| Total Confounder negative samples- from the Sero-Evaluation Unit (SEU), Manchester that are rheumatoid factor (12 samples), CMV (6 samples), EBV (19 samples) or VZV (13 samples) positive. All but one were negative using the Euroimmun IgG assay |
| 472 samples |
| 0 |
| 100% (99.1-100) |
| 100% (95.8-100.0) for Confounder and Porton sample sets (n=85) combined; results for individual sub-groups not reported individually |

| Roche Elecsys® |
| https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/887222/PHE_Evaluation_of_Roche_Elecsys_anti_SARS_CoV_2.pdf, PHE, 18/May/2020 |
| Porton negative samples- from the RIPL 2015 Lyme disease negative sample collection |
| 35 samples |
| 0 |
| Not reported individually |
| Siemens |
| No additional published data available at the time of review |
Table S4. Summary of external evaluations of assay sensitivity

| Publication DOI/URL, first author, date | Cohort | Sample number | Reactive/positive | Sensitivity (95% CI) | Notes |
|----------------------------------------|--------|---------------|------------------|----------------------|-------|
| 10.1093/clinchem/hvaa120 Tang MS et al, 18 Jun/2020 (as BioRxiv preprint on 10 May/2020) | Abbott | Post-symptom onset: | 12 specimens from 48 individuals | 0% (0.00, 26.47) | Multiple samples from individuals included, which may not accurately reflect intra-individual variation. Longitudinal samples from individuals used. |
| | | <3 days post-symptom onset | 6 | 20 | 30.0% (11.89, 54.28) |
| | | 3-7 days post-symptom onset | 23 | 11 | 47.8% (26.82, 69.41) |
| | | ≥14 days post-symptom onset | 48 | 45 | 93.8 (82.80, 98.69%) |
| | | Post-positive PCR: | 42 specimens from 23 individuals | 20 | 47.6% (32.0, 63.6) |
| | | <3 days post-positive PCR | 42 | 20 | 47.6% (32.0, 63.6) |
| | | 3-7 days post-positive PCR | 22 | 13 | 59.1% (36.3, 79.3) |
| | | 8-13 days post-positive PCR | 23 | 16 | 69.6% (47.1, 86.8) |
| | | ≥14 days post-positive PCR | 16 | 13 | 81.3% (54.4, 96.0) |
| 10.1128/JCM.00941-20, Bryan A et al, 7 May/2020 | RT-PCR positive, March-April 2020 | 680 specimens from 125 individuals | 53.1% (39.4, 66.3) | Proposed the use of AUC analysis to adjust thresholds: “These analyses indicated that optimal thresholds for the serologic diagnosis of SARS-CoV-2 was 1.42-1.49 at ≥ 17 days from symptom onset (sensitivity and specificity 100%); 0.7 at ≥ 14 days from onset (Sens 97.9%, Spec 99.6%); 0.7 at ≥ 10 days from onset (Sens 94.4%, Spec 99.6%); and 0.7 at ≥ 7 days from onset (Sens 88.0%, Spec 99.6%)” |
| | | Post-symptom onset: | ≤7 days post-symptom onset | 82.4% (51.0, 76.4) |
| | | 8-10 days post-symptom onset | 96.9% (89.5, 99.5) |
| | | 11-14 days post-symptom onset | 100% (95.1, 100) |
| | | 15-17 days post-symptom onset | 88.7% (78.5, 94.4) |
| | | Post-positive PCR: | ≤7 days post-positive PCR | 97.2% (90.4, 99.5) |
| | | 8-10 days post-positive PCR | 100.0% (95.4, 100.0) |
| | | 11-14 days post-positive PCR | 100.0% (95.5, 100.0) |
| | | 15-17 days post-positive PCR | 100.0% (95.5, 100.0) |
| https://doi.org/10.1101/2020.05.18.20101618, Jääskeläinen AJ et al, 22 May/2020 | RT-PCR positive by one of three methods: “...cobas® SARS-CoV-2 test on the Cobas® 6800 system (Roche Diagnostics, Basel, Switzerland), Amplidiag® COVID-19 test (Mobidiag, Espoo, Finland) and a protocol based on Corman et al (2020).” | 70 samples from 62 individuals | 80.5% (NA) | Longitudinal samples from the same individuals included, small sample sizes. |
| https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/928864/COVID-19_RTPCR_Positive_102220.pdf | RT-PCR positive, otherwise healthy individuals, “14 had an onset date ≤14 days prior to sample collection and 81 had an onset date ≥14 days prior to | 96 samples, unknown number of individuals | 80.5% (NA) | It should be noted here that none of the patients with previously positive PCR who tested negative by this assay had been |
| Sample Date | Asymptomatic | Hospital admission to sample date <=10 |
|-------------|--------------|--------------------------------------|
| 11-20 days  | 5            | 100% (47.8-100)                      |
| 21-30 days  | 31           | 93.6% (78.6-99.2)                    |
| 31-40 days  | 37           | 94.6% (81.8-99.3)                    |
| 41-50 days  | 8            | 87.5% (47.3-99.7)                    |
| From 14 days| 82           | 93.9% (86.3-98.0)                    |
| From 21 days| 76           | 93.4% (85.3-97.8)                    |

The conclusion states:

“93.4% (95%CI 85.3-97.8) for samples collected ≥14 days post symptom onset
93.9% (95%CI 86.3-98.0) for samples collected ≥21 days post symptom onset.”

DiAsorin Liaison SARS-CoV-2 S1/S2 IgG

| Study Reference                                                                 | Sample Description                                                                 | Sensitivity (%) | Specificity (%) |
|--------------------------------------------------------------------------------|-----------------------------------------------------------------------------------|-----------------|-----------------|
| 10.1515/cclm -2020-0594, Tré-Hardy M et al, 25/May/2020 | RT-PCR-positive patients with mild, moderate, severe and critical infection based on CT appearances and clinical symptoms | None provided | 91% (79, 96)    |
| https://doi.org/10.1101/2020.05.19.105445, Bonelli, F et al, 20/May/2020 | RT-PCR positive by one of three methods: -cobas® SARS-CoV-2 test on the Cobas® 6800 system (Roche Diagnostics, Basel, Switzerland), Amplidiag® COVID-19 test (Mobidiag, Espoo, Finland) and a protocol based on Corman et al (2020).“ | 43.8% (NA) | None provided |
| https://doi.org/10.1101/2020.05.18.20101618, Plebani M et al, 26/May/2020 | Microneutralisation antibody positive samples ≤5 days post RT-PCR-positive | None provided | 22.6% (14.2, 33.0) |
| https://doi.org/10.1101/2020.05.22.20106328, Jääskeläinen AJ et al, 22/May/2020 | 80 with at least one positive nasopharyngeal swab, consisting of: 16 healthcare workers, not hospitalised, SARS-CoV-2 positive | Sens 82.4% (71.2, 90.5) | Using optimised thresholds: Sens 97.1% (89.8, 99.6) |

Unknown number of individuals tested.
| 32 hospitalised SARS-CoV-2 patients with moderate disease (not requiring ventilation) | 32 hospitalised SARS-CoV-2 patients with severe disease (requiring ventilation) |
|---|---|
| 53 samples, number individuals unknown. | Number of individuals not stated, all were from patients with severe disease. |
| RT-PCR confirmed COVID-19 patients with different levels of disease severity | Included here as data different from product literature above. |
| All Severe | 41 samples |
| >14 days post symptoms | 40 |
| 73.58% (60.42, 83.56) | 97.6% (87.4, 99.6) |
| 94.44% (74.24, 99.72) | Different sensitivity reported from that of the manufacturer; provenance of the results reported here unclear. |

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| RT-PCR positive from a swab sample | Report symptom onset to sample date | 93 samples, number of individuals unknown |
|---|---|---|
| | | 78 |
| 11-20 days | 4 | 3 |
| 21-30 days | 35 | 28 |
| 31-40 days | 30 | 28 |
| 41-50 days | 8 | 8 |
| From 14 days | 77 | 67 |
| From 21 days | 73 | 64 |
| Hospital admission to sample date | 71.4% (41.9-91.6) | 50.0% (1.3-98.7) |
| <=10 days | 14 | 10 |
| Asymptomatic, not admitted | 2 | 1 |
| Number of individuals not reported. Small numbers in subgroups. | Number of individuals not reported. Small numbers in subgroups. |
### Table S5. Positive and negative predictive values (PPV, NPV) for each assay, using the manufacturer’s threshold and sensitivity and specificity for samples tested at ≥20 days, at population prevalences of 5%, 10%, 20% and 50%. Absolute numbers of false negative (FN) and false positive (FP) per million tests are also shown.

| Manufacturer | Population prevalence | Specificity (95% CI) | Sensitivity (95% CI) | FN per 1m tests (95% CI) | FP per 1m tests (95% CI) | Total errors per 1m tests (95% CI) | PPV | NPV |
|--------------|-----------------------|----------------------|----------------------|-------------------------|-------------------------|----------------------------------|-----|-----|
| Siemens      | 5%                    | 99.9 (99.4-100)      | 97.2 (90.2-94.8)     | 7300 (7134-7469)        | 900 (842-961)           | 15400 (15160-15643)               | 99.6%| 98.2%|
|              | 10%                   | 99.9 (99.4-100)      | 96.2 (90.2-94.8)     | 36500 (3533-3770)       | 950 (891-1012)          | 12350 (12135-12568)               | 98.0%| 99.6%|
|              | 20%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 1900 (1817-1987)        | 10450 (10252-10651)     | 12350 (12135-12568)               | 92.2%| 99.8%|
|              | 50%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 450 (409-494)           | 9500 (9311-962)         | 9950 (9756-10147)                 | 83.9%| 99.95%|
| Roche        | 5%                    | 99.9 (99.3-100)      | 97.2 (95.4-98.4)     | 1400 (1328-1475)        | 1900 (1816-1987)        | 3300 (3189-3414)                  | 96.2%| 99.85%|
|              | 10%                   | 99.9 (99.4-100)      | 98.1 (96.6-99.1)     | 950 (891-1012)          | 950 (891-1012)          | 1900 (1816-1987)                  | 98.1%| 99.90%|
|              | 20%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 900 (842-961)           | 9000 (8158-9187)        | 9900 (9707-10100)                 | 91.7%| 99.90%|
|              | 50%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 7600 (7431-7772)        | 9000 (8266-8985)        | 9800 (9608-9995)                  | 96.1%| 99.77%|
| Siemens      | 5%                    | 99.9 (99.4-100)      | 98.1 (96.6-99.1)     | 1900 (1816-1987)        | 900 (842-961)           | 2800 (2697-2906)                  | 99.1%| 99.79%|
|              | 10%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 3800 (3680-3923)        | 800 (746-857)           | 15400 (15160-15643)               | 99.6%| 98.2%|
|              | 20%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 1600 (1523-1680)        | 500 (457-546)           | 37000 (36631-37372)               | 99.9%| 93.2%|
|              | 50%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 8500 (8184-8877)        | 500 (457-546)           | 15000 (14763-15240)               | 99.9%| 99.10%|
| Abbotsyn     | 5%                    | 99.9 (99.4-100)      | 97.2 (90.2-94.8)     | 19000 (18733-19270)     | 5500 (5356-5647)        | 24500 (24198-24805)               | 98.9%| 96.30%|
|              | 10%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 45000 (4370-4653)       | 5000 (4863-5140)        | 5000 (9311-962)                   | 99.0%| 97.27%|
|              | 20%                   | 99.9 (99.4-100)      | 99.1 (97.8-99.7)     | 14000 (13771-14232)     | 1000 (939-1064)         | 15000 (14763-15240)               | 99.8%| 97.27%|
|              | 50%                   | 99.9 (99.4-100)      | 98.1 (96.6-99.1)     | 9500 (9311-962)         | 500 (457-546)           | 10000 (9806-10197)                | 99.9%| 98.13%|
Table S6. Summary of concordance/discordance of results between assays for “known positive” samples analysed as part of the main analysis. “+” denotes a positive result, “−” a negative result and “+/−” an equivocal result (the latter relevant for the DiaSorin assay only). These data represent last samples per patient.

| Abbott | DiaSorin | OIA | Roche | Siemens | n | Acute samples (<20 days post symptom onset) | % Acute | Samples from PCR-positive cases (≥20 days post symptom onset) | % Positive | Pre-pandemic negative | % Negative |
|--------|----------|-----|-------|---------|---|---------------------------------------------|---------|-------------------------------------------------|-----------|----------------------|-----------|
| -      | -        | -   | -     | -       | 977| 25                                         | 2.6     | 3                                               | 0.3       | 949                  | 97.1      |
| +      | +        | +   | +     | +       | 551| 62                                          | 11.3    | 489                                             | 88.7      | 0                    | 0.0       |
| -      | +        | -   | -     | -       | 19 | 5                                           | 26.3    | 4                                               | 21.1      | 10                   | 52.6      |
| -      | +        | +   | -     | -       | 18 | 2                                           | 11.1    | 16                                              | 88.9      | 0                    | 0.0       |
| +      | -        | +   | +     | +       | 12 | 6                                           | 50.0    | 6                                               | 50.0      | 0                    | 0.0       |
| -      | +        | -   | -     | -       | 11 | 0                                           | 0.0     | 0                                               | 0.0       | 11                   | 100.0     |
| -      | +        | +   | -     | -       | 10 | 4                                           | 40.0    | 6                                               | 60.0      | 0                    | 0.0       |
| -      | -        | +   | -     | -       | 5  | 2                                           | 40.0    | 1                                               | 20.0      | 2                    | 40.0      |
| -      | -        | -   | +     | -       | 5  | 4                                           | 80.0    | 1                                               | 20.0      | 0                    | 0.0       |
| -      | +/−      | +   | +     | +       | 5  | 1                                           | 20.0    | 4                                               | 80.0      | 0                    | 0.0       |
| +      | +/−      | +   | +     | -       | 4  | 3                                           | 75.0    | 1                                               | 25.0      | 0                    | 0.0       |
| +      | +/−      | +   | +     | +       | 4  | 1                                           | 25.0    | 3                                               | 75.0      | 0                    | 0.0       |
| -      | -        | +   | +     | +       | 3  | 1                                           | 33.3    | 2                                               | 66.7      | 0                    | 0.0       |
| -      | -        | -   | +     | +       | 2  | 0                                           | 0.0     | 1                                               | 50.0      | 1                    | 50.0      |
| -      | +/−      | -   | -     | -       | 2  | 0                                           | 0.0     | 0                                               | 0.0       | 2                    | 100.0     |
| +      | +/−      | +   | +     | -       | 2  | 2                                           | 100.0   | 0                                               | 0.0       | 0                    | 0.0       |
| -      | +/−      | +   | +     | -       | 1  | 0                                           | 0.0     | 1                                               | 100.0     | 0                    | 0.0       |
| +      | +/−      | +   | +     | +       | 1  | 1                                           | 100.0   | 0                                               | 0.0       | 0                    | 0.0       |
| +      | +/−      | +   | +     | -       | 1  | 0                                           | 0.0     | 0                                               | 0.0       | 1                    | 100.0     |
| +      | +/−      | +   | +     | +       | 1  | 1                                           | 100.0   | 0                                               | 0.0       | 0                    | 0.0       |
| +      | +/−      | +   | +     | -       | 1  | 1                                           | 100.0   | 0                                               | 0.0       | 0                    | 0.0       |
Table S7. Summary of concordance/discordance of results between assays for early acute samples analysed at <14 days post-symptom onset. First sample per patient analysed; total n=118.

|       | Abbott | DiaSorin | OIA | Roche | Siemens |
|-------|--------|----------|-----|-------|---------|
| % of  | -      | -        | -   | -     | -       |
| samples| 32     | 27       | 40  | 34    | 11      |
| +     | +      | +        | +   | +     | +       |
|       | 6      | 5        | 3   | 3     | 9       |
| -     | -      | -        | -   | -     | -       |
|       | 0      | 0        | 0   | 0     | 0       |
| %     | -      | -        | -   | -     | -       |
| 2     | 1.7    | 4        | 0.8 | 0.8   | 0.8     |
| +     | +      | +        | +   | +     | +       |
|       | 3      | 3        | 0   | 0     | 0       |
| -     | -      | -        | -   | -     | -       |
|       | 0      | 0        | 0   | 0     | 0       |
| %     | +      | +        | +   | +     | +       |
| 1     | 0.8    | 1        | 0.8 | 0.8   | 0.8     |
| +     | +      | -        | -   | -     | -       |
|       | 2      | 1.7      | 0   | 0     | 0       |
| +     | +      | +        | +   | +     | +       |
|       | 1      | 0.8      | 0   | 0     | 0       |
| +     | +      | +        | +   | +     | +       |
|       | 1      | 0.8      | 0   | 0     | 0       |

Acute samples (<14 days post symptom onset)
**Figure S1. Sample collections and inclusions/exclusions.** For de-duplication of samples by individual, the latest sample meeting the MHRA criteria (i.e. latest sample taken ≥20 post-symptom onset) was included. The Table below the figure summarises the partial results for the five samples that were of insufficient volume to run across all platforms, and the text below the table the results for the 18 samples that did not pass QC due to liquid handling failures for the OIA.

| Sample barcode | Expected result | Days since symptom onset | Platform | Actual result |
|----------------|----------------|--------------------------|----------|---------------|
| 900753         | Negative       | n/a                      | Abbott   | Negative      |
|                |                |                          | DiaSorin | Negative      |
|                |                |                          | Roche    | Negative      |
| 500379         | Positive       | 40                       | Abbott   | Positive      |
|                |                |                          | DiaSorin | Positive      |
| 500380         | Positive       | 41                       | Abbott   | Positive      |
|                |                |                          | DiaSorin | Positive      |
| 500381         | Positive       | 41                       | Abbott   | Negative      |
|                |                |                          | DiaSorin | Negative      |
| 500384         | Positive       | 42                       | Abbott   | Positive      |
|                |                |                          | DiaSorin | Positive      |
For the 18 samples that failed on the OIA, all were in the pre-pandemic (known negative) sample group, and all were negative by Abbott, Roche and Siemens; a single sample was positive on the DiaSorin assay (900079).
Figure S2. Sensitivity and specificity (95% confidence intervals) plotted for each assay on all samples ≥20 days post-symptom onset in confirmed laboratory cases of SARS-CoV-2 for positive cases, and >6 months prior to the first known COVID-19 cases for negatives. A target performance in line with the UK MHRA Target Product Profile is shown (dashed line) including the required lower bound of the 95% confidence interval (dotted line) for both sensitivity and specificity. Data are presented for all known negative samples (Abbott n=995, DiaSorin n=995, OIA n=977, Roche n=995, Siemens n=994) and all known positive samples run across (Abbott n=540, DiaSorin n=540, OIA n=540, Roche n=536, Siemens n=536) assays; equivocal results were excluded from the calculation of sensitivity and specificity for the DiaSorin assay (n=9).
Figure S3. Distribution of numerical results obtained for each commercial assay on all samples taken ≥20 days post-symptom onset (i.e. not restricted to samples only run across all platforms). Results are represented as A. histograms, to enable assessment of the frequency of values, and B. dotplots, to review scatter of values, especially around thresholds. Pre-specified assay thresholds are shown as dashed lines. For the purposes of plotting values on a log scale, values of zero were set to the lowest non-zero value and results of greater or less than the largest or smallest values were truncated to the largest and smallest values. Data are presented for all known negative samples (Abbott n=995, DiaSorin n=995, OIA n=977, Roche n=995, Siemens n=994) and all known positive samples run across (Abbott n=540, DiaSorin n=540, OIA n=540, Roche n=536, Siemens n=536) assays.
Figure S4. Sensitivity and specificity (95% confidence intervals) plotted for each assay on samples ≥14 days post-symptom onset in confirmed laboratory cases of SARS-CoV-2 for positive cases, and >6 months prior to the first known COVID-19 cases for negatives. A target performance in line with the UK MHRA Target Product Profile is shown (dashed line) including the required lower bound of the 95% confidence interval (dotted line) for both sensitivity and specificity. Data are presented for samples run across all platforms (n=976 and 561 for pre-pandemic and samples from positive cases respectively).
Figure S5. Sensitivity and specificity (95% confidence intervals) plotted for each assay on samples ≥30 days post-symptom onset in confirmed laboratory cases of SARS-CoV-2 for positive cases, and >6 months prior to the first known COVID-19 cases for negatives. A target performance in line with the UK MHRA Target Product Profile is shown (dashed line) including the required lower bound of the 95% confidence interval (dotted line) for both sensitivity and specificity. Data are presented for samples run across all platforms (n=976 and n=490 for pre-pandemic and samples from positive cases respectively).
Figure S6. ROC curves for each assay on samples taken ≥14 days after the onset of symptoms. The green shaded area represents sensitivity and specificity of ≥98% and ≥98% respectively. Assay values associated with 10 exemplar points on the ROC curve are shown in each panel. Data are presented for 976 known negative samples and 561 known positive samples run on each assay.
Figure S7. ROC curves for each assay on samples taken ≥30 days after the onset of symptoms. The green shaded area represents sensitivity and specificity of ≥98% and ≥98% respectively. Assay values associated with 10 exemplar points on the ROC curve are shown in each panel. Data are presented for 976 known negative samples and 490 known positive samples run on each assay.
Figure S8. Sensitivity and specificity (95% confidence intervals) plotted for each assay with alternative assay thresholds to keep specificity $\geq 98\%$ and revised criteria to show samples $\geq 30$ days after the appearance of first symptoms. For each assay the lowest threshold that kept specificity $\geq 98\%$ was chosen (Abbott=0.49, DiaSorin=10, OIA=7.19 million normalised standard units, Roche=0.128, Siemens=0.29). The UK MHRA target performance is shown (dashed line) including the required lower bound of the 95% confidence interval (dotted line) for both sensitivity and specificity. Data are presented for 976 known negative samples and 490 known positive samples run on each assay.
Figure S9. Values for 8-point 1:2 dilution series of high-volume plasma controls defined as having high, medium and low titre antibodies (by EUROIMMUN; ratio values of 33·33, 4·34 and 2·50 respectively, where ratio is the optical density of the sample divided by the optical density of the calibrator [ratio of $\geq 1.1$ is positive]; the S1 component of the SARS-CoV-2 spike protein is the antigen target for the EUROIMMUN assay), and a known pre-pandemic negative control. The “high” dilution series is represented by samples QC1001-QC1008, the “medium” dilution series by QC1009-1016, and the “low” dilution series by samples QC1017-1024. QC1025 is the negative control (sample BD01). Values were log(2) transformed prior to plotting.
Figure S10. Percentage of tests from SARS-CoV-2 RT-PCR-positive individuals positive over time by serology platform. Samples from <20 days from symptom onset, excluded from the main analysis, are included here. Panel A shows the percentage by time since symptom onset and panel B the percentage by the time since the individual’s first positive RT-PCR test.
Figure S11. Modelled antibody trajectories by assay, by day post-symptom onset. First sample per patient only were included. The black line shows the modelled mean and the grey shading the 95% confidence interval for the mean.
Figure S12. Sensitivity for each assay by disease severity (asymptomatic, mild, severe, critical/death; n=158). Disease severity was defined in line with WHO guidance(2) as follows: asymptomatic = no symptoms; mild = no oxygen requirement; severe = SaO2 ≤93%; critical = respiratory failure requiring intubation. Severity category was assigned on the day of sampling (asymptomatic n=13, mild n=122, severe n=16, critical/death n=7).
### STARD checklist for evaluations of diagnostic accuracy

| Section & Topic | No | Item                                                                 | Reported on page # |
|-----------------|----|----------------------------------------------------------------------|--------------------|
| **Title or Abstract** |    | Identification as a study of diagnostic accuracy using at least one measure of accuracy (such as sensitivity, specificity, predictive values, or AUC) | Title, Abstract    |
| **Abstract**    | 2  | Structured summary of study design, methods, results, and conclusions (for specific guidance, see STARD for Abstracts) | Abstract            |
| **Introduction**| 3  | Scientific and clinical background, including the intended use and clinical role of the index test | Introduction        |
|                 | 4  | Study objectives and hypotheses                                      | Introduction        |
| **Methods**     |    |                                                                      |                    |
| **Study design**| 5  | Whether data collection was planned before the index test and reference standard were performed (prospective study) or after (retrospective study) | Methods - study design |
| **Participants**| 6  | Eligibility criteria                                                 | Methods - study design |
|                 | 7  | On what basis potentially eligible participants were identified (such as symptoms, results from previous tests, inclusion in registry) | Methods - study design |
|                 | 8  | Where and when potentially eligible participants were identified (setting, location and dates) | Methods - study design, Appendix - Table S1 |
|                 | 9  | Whether participants formed a consecutive, random or convenience series | Methods - study design |
| **Test methods**|    |                                                                      |                    |
| **10a**         |    | Index test, in sufficient detail to allow replication                | Methods - study design and procedures, Appendix - supplementary methods section C, previous publication |
| **10b**         |    | Reference standard, in sufficient detail to allow replication        | Methods - study design |
| **11**          |    | Rationale for choosing the reference standard (if alternatives exist) | Methods - study design |
| **12a**         |    | Definition of and rationale for test positivity cut-offs or result categories of the index test, distinguishing pre-specified from exploratory | Methods - study design, Appendix - Table S1, Appendix - supplementary methods section C |
| **12b**         |    | Definition of and rationale for test positivity cut-offs or result categories of the reference standard, distinguishing pre-specified from exploratory | Methods - study design, Appendix - Table S1, Appendix - supplementary methods section C |
| **13a**         |    | Whether clinical information and reference standard results were available to the performers/readers of the index test | Methods - procedures |
| **13b**         |    | Whether clinical information and index test results were available to the assessors of the reference standard | Methods - procedures |
| **Analysis**    | 14 | Methods for estimating or comparing measures of diagnostic accuracy  | Methods - study design, procedures, outcomes and statistical analysis |
| 15 | How indeterminate index test or reference standard results were handled | Methods - study design, procedures, outcomes and statistical analysis |
| 16 | How missing data on the index test and reference standard were handled | Methods - study design, procedures, outcomes and statistical analysis, Appendix Figure S1 |
| 17 | Any analyses of variability in diagnostic accuracy, distinguishing pre-specified from exploratory | Methods - study design, procedures, outcomes and statistical analysis |
| 18 | Intended sample size and how it was determined | Methods - outcomes and statistical analysis |

**RESULTS**

**Participants**

| 19 | Flow of participants, using a diagram | Appendix Figure S1 |
| 20 | Baseline demographic and clinical characteristics of participants | Appendix Table S1 |
| 21a | Distribution of severity of disease in those with the target condition | Appendix Fig S12, dataset available at https://doi.org/10.6084/m9.figshare.c.5046032.v1 |
| 21b | Distribution of alternative diagnoses in those without the target condition | No detail available, mentioned in Discussion |
| 22 | Time interval and any clinical interventions between index test and reference standard | Methods - study design, Appendix Table S1 |

**Test results**

| 23 | Cross tabulation of the index test results (or their distribution) by the results of the reference standard | Table 1 |
| 24 | Estimates of diagnostic accuracy and their precision (such as 95% confidence intervals) | Results, Table 1, Fig 1, Fig 2, Appendix Table S5-S7, Fig S2-S8, S10, S12 |
| 25 | Any adverse events from performing the index test or the reference standard | Not applicable |

**DISCUSSION**

| 26 | Study limitations, including sources of potential bias, statistical uncertainty, and generalisability | Discussion |
| 27 | Implications for practice, including the intended use and clinical role of the index test | Abstract, Discussion |

**OTHER INFORMATION**

| 28 | Registration number and name of registry | Not applicable |
| 29 | Where the full study protocol can be accessed | https://doi.org/10.6084/m9.figshare.c.5046032.v1 |
| 30 | Sources of funding and other support, role of funders | Abstract, Funding section |
# PRISMA checklist for systematic review

## PRISMA 2009 Checklist

| Section/Topic                      | # | Checklist item                                                                                                                                                                                                 | Reported on page # |
|------------------------------------|---|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------|
| **TITLE**                          |   |                                                                                                                                                                                                               |                    |
| Title                              | 1 | Identify the report as a systematic review, meta-analysis, or both.                                                                                                                                              | Abstract, Research in context |
| **ABSTRACT**                       |   |                                                                                                                                                                                                               |                    |
| Structured summary                 | 2 | Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number. | Abstract, Research in context, Appendix Supplementary Methods section A |
| **INTRODUCTION**                   |   |                                                                                                                                                                                                               |                    |
| Rationale                          | 3 | Describe the rationale for the review in the context of what is already known.                                                                                                                                   | Research in context |
| Objectives                         | 4 | Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).                                                        | Appendix Supplementary Methods section A |
| **METHODS**                        |   |                                                                                                                                                                                                               |                    |
| Protocol and registration          | 5 | Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address) and, if available, provide registration information including registration number.                                             | No protocol registered for systematic review |
| Eligibility criteria               | 6 | Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.                         | Supplementary Methods section A |
| Information sources                | 7 | Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.                                                | Research in context, Supplementary Methods |
| Search                             | 8 | Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.                                                                              | Section A          |
| Study selection                    | 9 | State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).                                                   | Supplementary Methods section A |
| Data collection process            | 10| Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.                                     | Supplementary Methods section A |
| Data items                         | 11| List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.                                                                             | Supplementary Methods section A |
| Risk of bias in individual studies | 12| Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.       | Not evaluated       |
| Summary measures                   | 13| State the principal summary measures (e.g., risk ratio, difference in means).                                                                                                                                     | Supplementary Methods section A |
| Synthesis of results               | 14| Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., I²) for each meta-analysis.                                                           | Not applicable      |

## RESULTS

| Section/Topic                      | # | Checklist item                                                                                                                                                                                                 | Reported on page # |
|------------------------------------|---|---------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|--------------------|
| Risk of bias across studies       | 15| Specify any assessment of risk of bias that may affect the cumulative evidence (e.g., publication bias, selective reporting within studies).                                                              | Not evaluated      |
| Additional analyses               | 16| Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.                                                              | Not applicable      |
| Study selection                   | 17| Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.                                               | Research in context |
| Study characteristics | 18 | For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations. | Supplementary Methods section A |
|-----------------------|----|-----------------------------------------------------------------|---------------------------------|
| Risk of bias within studies | 19 | Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12). | Not evaluated |
| Results of individual studies | 20 | For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot. | Not applicable |
| Synthesis of results | 21 | Present results of each meta-analysis done, including confidence intervals and measures of consistency. | Not applicable |
| Risk of bias across studies | 22 | Present results of any assessment of risk of bias across studies (see Item 15). | Not evaluated |
| Additional analysis | 23 | Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression (see item 16)). | Not applicable |

**DISCUSSION**

| Summary of evidence | 24 | Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers). | Research in context |
| Limitations | 25 | Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias). | Supplementary Tables S3, S4 |
| Conclusions | 26 | Provide a general interpretation of the results In the context of other evidence, and implications for future research. | Research in Context section |

**FUNDING**

| Funding | 27 | Describe sources of funding for the systematic review and other support (e.g., supply of data), role of funders for the systematic review. | Funding section in Abstract and at end of manuscript |
Supplementary References
1. Emmenegger M, de Cecco E, Lamparter D, et al. Early plateau of SARS-CoV-2 seroprevalence identified by tripartite immunoassay in a large population. medRxiv 2020. DOI: https://doi.org/10.1101/2020.05.31.20118554.
2. Word Health Organisation (WHO). Report of the WHO-China Joint Mission on Coronavirus Disease 2019 (COVID-19). 2020. https://www.who.int/docs/default-source/coronaviruse/who-china-joint-mission-on-covid-19-final-report.pdf. Accessed: 18/Jun/2020.