Philippine COVID-19 Living Clinical Practice Guidelines
Institute of Clinical Epidemiology, National Institutes of Health, UP Manila
In cooperation with the Philippine Society for Microbiology and Infectious Diseases
Funded by the Department of Health

EVIDENCE SUMMARY

RESEARCH QUESTION: **Among patients suspected to have COVID-19, should antibody tests be used to diagnose COVID-19 among vaccinated adults and children?**

Review by: Jhon Ryan Enriquez, MD, FPCP, DPSMID, Michelle Cristine Miranda, MD, Maria Teresa Sanchez-Tolosa, MD, D Clin Epi, Evalyn A. Roxas, MD, MPH, Donna Isabel S. Capili, MD, Marissa M. Alejandria, MD, MSc

RECOMMENDATIONS

Recommendations	Certainty of Evidence	Strength of Recommendation
There is no evidence to recommend for or against antibody testing to diagnose COVID-19 disease among vaccinated patients.	None	None
We suggest against the routine measurement of SARS-CoV-2 antibody titers after vaccination. In the rare situations where we need to determine prior COVID-19 disease or infection, we suggest the use of nucleocapsid antibody testing among vaccinated individuals, along with infectious disease specialist consultation.	Very low	Weak

Consensus Issues

The panel noted that there are no available studies on the use of antibody tests for the diagnosis of COVID-19. The panel also brought up issues of cost and goals in antibody testing, noting that useful clinical applications of antibody testing are limited such as in MIS-C cases and vaccination in immunocompromised individuals, hence the emphasis for expert consultation in the use of these tests.

KEY FINDINGS

- We included six (6) seroprevalence studies (n=24,070 samples) that investigated the diagnostic accuracy of antibody tests in the detection of past exposure to COVID-19 using reverse transcription polymerase chain reaction (RT-PCR) as the reference standard.
- The overall certainty of evidence is very low due to serious risk of bias, very serious inconsistency and very serious indirectness.
- The pooled sensitivity of antibody tests was 99% (95% CI 96.7–99.7; I^2=73.2%; very low certainty) while pooled specificity was 11.9% (95% CI 2.5-41.4; I^2=73.2%; very low certainty). Heterogeneity across the studies was substantial (I^2=0-99%).
- Sensitivity was high across all subgroups ranging from 95%-100%, while specificity was noted to be high only in a subgroup measuring Nucleocapsid antibodies (90% specificity; 90% CI 89%- 91%; very low certainty).
- No studies evaluating the accuracy of antibody tests compared to RT-PCR in determining COVID-19 disease were found.
- No direct studies evaluating the accuracy of antibody tests compared to RT-PCR in determining past COVID-19 exposure were found. Only seroprevalence studies with subgroups of interest that allow the construction of a 2x2 table for diagnostic accuracy were included.

INTRODUCTION

In SARS-CoV-2, the structural nucleocapsid and spike proteins were found to be dominant antigens for host immune response and have become the basis for detecting antibodies to immunoglobulins (Ig) binding to these proteins [1]. Generally, antibodies are classified as neutralizing antibodies, i.e., cause virus particles to lose infectivity, and binding antibodies [2].

While not a replacement for virologic testing, SARS-CoV-2 serology can be useful in clinical, occupational health and public health settings [3]. Validating antibody tests is important because certain assays may cross-react with other coronaviruses, among other concerns. Antibody tests should have high sensitivity and specificity to be clinically useful; specificity is particularly important in large serosurveillance studies in areas with a low expected prevalence of prior SARS-CoV-2 infection [3].

Before broad vaccination campaigns, serosurveys were relatively straightforward to interpret because SARS-CoV-2 antibodies were elicited primarily by symptomatic or asymptomatic infection. SARS-CoV-2 humoral immune responses are commonly characterized by detection of antibodies against the spike glycoprotein (S), the receptor-binding domain (RBD) within the spike glycoprotein, and the nucleocapsid protein (N). Infection elicits antibodies against all three targets; however, many vaccines (eg, current mRNA and viral vector vaccines) only target S protein–derived antigens. Hence, it is not possible to distinguish infection- and vaccine-induced immunity via serostatus alone. It is now important to differentiate seropositivity due to past infection, past vaccination, or both. This stratification enables public health officials to monitor true infection rates, and in turn estimate infection fatality ratios (IFRs) and asymptomatic infections [4].

Distinguished groups released current guidelines in terms of the utility of Serology as a diagnostic tool for COVID-19 infection, However, these studies only included unvaccinated individuals [5,6].

As of April 2023, the Philippine Food and Drug Administration (FDA) lists 38 approved antibody immunoassays and 14 rapid antibody test kits after performance validation by the Research Institute for Tropical Medicine (RITM) [7].

This review sought to answer the following clinical questions:

1. Should antibody tests be used to diagnose COVID-19 disease among vaccinated adults and children, using RT-PCR as a reference standard?
2. Should antibody tests be used to determine past COVID-19 exposure among vaccinated adults and children, using RT-PCR as a reference standard?

REVIEW METHODS

To identify possible studies, we ran an electronic search on several databases (PubMed, Cochrane CENTRAL, medRxiv, bioRxiv, ClinicalTrials.gov). Initial search was done March 1, 2021 to March 27, 2023 using the following terms: antibody test, antibody, serology, serologic test, Immunoglobulin, IgG, IgM, IgA, accuracy, sensitivity, specificity, predictive value, COVID-19, SARS-CoV-2, Corona virus, vaccinated, post vaccination. Observational studies, reviews, systematic review and meta-analysis were all included. A filter date of March 1, 2021 onwards was used to include only studies published after the global rollout of vaccination which occurred last December 2020. The search did not yield any result that would answer our clinical question.

An augmented search, on the same databases and following the same filter, was done to check for seroprevalence studies that might have included subgroups of the desired population and outcome. We used the following MeSH terms: seroprevalence, serosurveillance, antibody, antibody test, serology, serologic test, vaccinated, post vaccination.

Studies that used SARS-CoV-2 antibody tests to determine COVID-19 seroprevalence among adults and children were included. A positive nucleic acid amplification test (NAAT) such as reverse transcription polymerase chain reaction (RT-PCR) was used as an acceptable reference standard following several previous reviews [5,6].

We excluded studies with target subgroups insufficient to construct a 2x2 table for diagnostic accuracy such as those who used quantitative measure of antibodies.

Methodological quality of included studies was assessed using QUADAS-2 via Revman version 5.4.1.The Meta-DiSc 2.0 was used for pooled studies and Revman V5.4.1 for single studies to determine sensitivity, specificity, and measures of heterogeneity as well as generate forest plots. Random effects models were used in conducting univariate meta-analyses for pooled studies of 3 and below and bivariate meta-analyses for pooled studies of 4 and above. Subgroup analyses according to the type of population tested, antibodies detected (e.g., IgG, IgM), specific antibody measured (e.g., anti-S, anti-M) and reference standard (e.g., self-reported, laboratory-confirmed) were performed.

RESULTS

Characteristics of included studies

Six seroprevalence studies to determine prior COVID-19 exposure using antibodies were included in this review. There was a total of seven comparisons of antibody tests with RT-PCR (n=24,070 samples). Seropositivity was as low as 17.7% to as high as 91% [12,13].

IgG antibodies was the most commonly measured antibody type among the studies (four studies) [8-10,12] Kanamori et al. measured total antibodies and Cerbino-neto et al. measured both IgG and IgM antibodies [11,13].

IgG Antibodies specific against spike and nucleocapsid protein was the most commonly measured (three studies) [8, 9, 11]. One study, that of Kanamori et al. measured specific antibodies against nucleocapsid protein [13]. Two studies measured anti-spike protein antibodies [10, 12].

Four of the six studies clearly defined a predetermined cut-off value for test positivity for the index test [8-10, 13].

Methodological Quality/Certainty of evidence

Three studies were at risk of recall bias because of self-reporting of the reference standard results [8, 11, 12]. One study also had the reference standard (RT-PCR) combined with a previously positive antibody test as the confirmation of a past COVID-19 exposure [10]. All of the studies' negative control group was based on not having been tested with the reference standard or not having symptoms of COVID-19 (hence, not tested for RT-PCR) instead of having a negative reference standard test. Moreover, the population being tested (vaccinated participants) for all of the studies had a tendency to have a positive index test, regardless of the results of the reference standard. Hence, all of the studies were deemed with high risk of bias.

Diagnostic Accuracy

The measures of diagnostic test accuracy varied across included studies (n=24,070 samples). The pooled sensitivity of antibody tests was 99% (95% CI 96.7–99.7; I^2=73.2%) while pooled specificity was 11.9% (95% CI 2.5-41.4; I^2=73.2%).

Heterogeneity was substantial (I^2=73.2%) and was not reduced in almost all subgroup analyses, except for subgroup measuring antibodies specific for spike protein, subgroup using a laboratory confirmed SARS-CoV-2 RT-PCR as a reference standard, and subgroup done amongst healthcare workers as the population (I^2=0%).

Sensitivity was highest in the study that detected IgG antibodies against the spike protein (n=14,483): 100% (95% CI 100-100) [12]. This was followed by the study that detected IgG antibodies against at least one antigen — NCP, RBD, or S2 (n=168): 100% (95% CI 100-100) [9]. Pooled studies that detected IgG Antibodies against the Spike protein showed very high sensitivity (n=14,669): 99.7% (CI 99.6-99.8%; I^2= 0%) [10, 12].

Meanwhile, specificity was highest in one study that detected antibodies against nucleocapsid protein (n= 3,708): 90% (95% CI 89-91) [13]. This same study reported a sensitivity of 96% (95%CI 94-98).

In pooled studies among adult populations, the sensitivity of antibody test was 99% (95% CI 97-100; I^2= 93.8%) while specificity was 12% (95% CI 1-61; I^2=99.9%) [9-10, 12-13]. In pooled studies among adult and children the sensitivity of antibody test was 95% (95% CI 94-96; I^2=62.3%) while specificity was 13% (95% CI 10-16; I^2=88.6%) [8, 11].

In pooled studies among population who used self-reported RT-PCR as a reference standard, the sensitivity of antibody test was 99% (95% CI 95-100; I^2=98.4%) while specificity was 8% (95% CI 3-17; I^2=99.6) [8, 11, 12]. In pooled studies among population who used Laboratory confirmed RT-PCR as a reference standard, the sensitivity of antibody test was 97% (95% CI 95-98%; I^2=0%) while specificity was 35% (95% CI 1-97; I^2=98.9%) [9, 13].

The overall certainty of evidence was rated Very Low due to serious risk of bias, very serious inconsistency and very serious indirectness.

RECOMMENDATIONS FROM OTHER GROUPS

The table below summarizes the recommendations from various regulatory agencies

Guideline / Agency	Recommendation	Strength of Recommendation / Certainty of Evidence
Philippine Food and Drug Administration (2021) [16]	There are **no currently available** FDA approved COVID-19 test kits in the Philippines that differentiate the antibody protection gained from natural COVID-19 infection and the immunity from vaccination.	-
Health Technology Assessment Council (Philippines) (2020) [17]	TAC does **not recommend** the use of rapid antibody tests… “in seroprevalence surveys, return-to-work decisions, or entry-to-country/ province policies due to the lack of evidence regarding the link of presence of antibodies and the immunity to subsequent infection AND on the persistence of protection from COVID-19.” *Non vaccinated population	-

European Centre for Disease Prevention and Control (2022) [18]	Serological **tests cannot be used to detect current infection with SARS-CoV-2**. These tests may be used to confirm a past infection with SARS-CoV-2 in clinical care settings or for public health purposes. Antibodies against the spike protein also become detectable after vaccination against SARS-CoV-2; therefore, attribution of positive serological tests to previous infections should be done with caution due to high vaccine coverage within the general population	
Infectious Disease Society of America (2020) [19] *Non vaccinated population	IDSA panel **suggests against using serologic testing to diagnose SARS-CoV-2** infection during the first two weeks (14 days) following symptom onset	Conditional recommendation, Very low certainty of evidence
	"The IDSA panel makes **no recommendation** either for or against using IgM antibodies to detect evidence of past SARS-CoV-2 infection"	
	"The IDSA panel suggests **against using IgA** antibodies to detect evidence of past SARS-CoV-2 infection"	
	"The IDSA panel suggests **against using IgM or IgG** antibody combination tests to detect evidence of past SARS-CoV-2 infection"	

ONGOING STUDIES AND RESEARCH GAPS

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ADDITIONAL CONSIDERATIONS FOR EVIDENCE TO DECISION (ETD) PHASE

Rapid antibody tests for COVID-19 may cost around ₱1,500 to 2,500 per test [14,15]. We found no evidence on the cost-effectiveness and acceptability of antibody tests in the Philippine setting.

REFERENCES

[1] Pecora ND, Zand MS. Measuring the Serologic Response to Severe Acute Respiratory Syndrome Coronavirus 2: Methods and Meaning. Clin Lab Med. 2020;40(4):603–14.

[2] Hanson KE, Caliendo AM, Arias CA, Englund JA, Hayden MK, Lee MJ, et al. Infectious Diseases Society of America Guidelines on the Diagnosis of COVID-19: Serologic Testing.

[3] Centers for Disease Control and Prevention. Interim Guidelines for COVID-19 Antibody Testing in Clinical and Public Health Settings. Cent Dis Control Prev [Internet]. 2020;1–8. Available from: https://www.cdc.gov/coronavirus/2019-ncov/lab/resources/antibody-tests-guidelines.html

[4] Duarte N, Yanes-Lane M, Arora RK, Bobrovitz N, Liu M, Bego MG, Yan T, Cao C, Gurry C, Hankins CA, Cheng MP, Gingras AC, Mazer BD, Papenburg J, Langlois MA. Adapting Serosurveys for the SARS-CoV-2 Vaccine Era. Open Forum Infect Dis. 2021 Dec 23;9(2):ofab632. doi: 10.1093/ofid/ofab632. PMID: 35103246; PMCID: PMC8755308.

[5] Fox T, Geppert J, Dinnes J, Scandrett K, Bigio J, Sulis G, Hettiarachchi D, Mathangasinghe Y, Weeratunga P, Wickramasinghe D, Bergman H, Buckley BS, Probyn K, Sguassero Y, Davenport C, Cunningham J, Dittrich S, Emperador D, Hooft L, Leeflang MM, McInnes MD, Spijker R, Struyf T, Van den Bruel A, Verbakel JY, Takwoingi Y, Taylor-Phillips S, Deeks JJ; Cochrane COVID-19 Diagnostic Test Accuracy Group. Antibody tests for identification of current and past infection with SARS-CoV-2. Cochrane Database Syst Rev. 2022 Nov 17;11(11):CD013652. doi: 10.1002/14651858.CD013652.pub2. PMID: 36394900; PMCID: PMC9671206.

[6] https://www.idsociety.org/practice-guideline/covid-19-guideline-serology

[7] (FOR PUBLIC VIEWING) 2021 APPROVED COVID-19 TEST KIT WITH RITM RESULTS as of December 9, 2022 https://docs.google.com/spreadsheets/d/13S12QST9RKlyX1ipUhqhmngwq3rdtbLRnDWPg8-OSqo/edit#gid=1687242007

[8] Murhekar MV, Bhatnagar T, Thangaraj JWV, Saravanakumar V, Santhosh Kumar M, Selvaraju S, Rade K, Kumar CPG, Sabarinathan R, Asthana S, Balachandar R, Bangar SD, Bansal AK, Bhat J, Chakraborty D, Chopra V, Das D, Devi KR, Dwivedi GR, Jain A, Khan SMS, Kumar MS, Laxmaiah A, Madhukar M, Mahapatra A, Ramesh T, Rangaraju C, Turuk J, Yadav S, Bhargava B; ICMR serosurveillance group. Seroprevalence of IgG antibodies against SARS-CoV-2 among the general population and healthcare workers in India, June-July 2021: A population-based cross-sectional study. PLoS Med. 2021 Dec 10;18(12):e1003877. doi: 10.1371/journal.pmed.1003877. PMID: 34890407; PMCID: PMC8726494

[9] Błaszczuk A, Michalski A, Malm M, Drop B, Polz-Dacewicz M. Antibodies to NCP, RBD and S2 SARS-CoV-2 in Vaccinated and Unvaccinated Healthcare Workers. Vaccines (Basel). 2022 Jul 22;10(8):1169. doi: 10.3390/vaccines10081169. PMID: 35893818; PMCID: PMC9329710

[10] Decarreaux D, Sevila J, Masse S, Capai L, Fourié T, Villarroel PMS, Amroun A, Nurtop E, Vareille M, Blanchon T, de Lamballerie X, Charrel R, Falchi A. Eight Months of Serological Follow-Up of Anti-SARS-CoV-2 Antibodies in France: A Study among an Adult Population. Int J Environ Res Public Health. 2022 Nov 18;19(22):15257. doi: 10.3390/ijerph192215257. PMID: 36429974; PMCID: PMC9691066.

[11] Cerbino-Neto J, Peres IT, Varela MC, Brandão LGP, de Matos JA, Pinto LF, da Costa MD, Garcia MHO, Soranz D, Maia MLS, Krieger MA, da Cunha RV, Camacho LAB, Ranzani O, Hamacher S, Bozza FA, Penna GO. Seroepidemiology of SARS-CoV-2 on a partially vaccinated island in Brazil: Determinants of infection and vaccine response. Front Public Health. 2022 Nov 14;10:1017337. doi: 10.3389/fpubh.2022.1017337. PMID: 36457326; PMCID: PMC9706255.

[12] Thon V, Piler P, Pavlík T, Andrýsková L, Doležel K, Kostka D, Pikhart H, Bobák M, Klánová J. Investigation of SARS-CoV-2 seroprevalence in relation to natural infection and vaccination between October 2020 and September 2021 in the Czech Republic: a prospective national cohort study. BMJ Open. 2023 Mar 10;13(3):e068258. doi: 10.1136/bmjopen-2022-068258. PMID: 36898746; PMCID: PMC10008433.

[13] Kanamori R, Yan Y, Ito K, Fukuda H, Hori S, Yamamoto T, Igawa G, Saito K, Horiuchi Y, Nojiri S, Nishizaki Y, Tabe Y, Takahashi K, Naito T. Increased SARS-CoV-2 seroprevalence and spread of infection without awareness among healthcare workers through 2020-2022 in a Japanese medical

center. Sci Rep. 2023 Mar 27;13(1):4941. doi: 10.1038/s41598-023-32193-4. PMID: 36973531; PMCID: PMC10040914.

[14] https://www.moneymax.ph/personal-finance/articles/swab-rapid-test-price

[15] https://medicalpinas.com/hi-precision-diagnostic-center-list-of-services-and-prices/#Blood_Test_Price

[16] https://www.fda.gov.ph/fda-advisory-no-2021-1270-use-of-covid-19-antibody-test-kits-for-assessment-of-immunity-after-covid-19-vaccination/

[17] https://hta.doh.gov.ph/2020/07/30/rapid-antibody-tests-ratsmarch-2020-assessment/

[18] https://www.ecdc.europa.eu/en/covid-19/latest-evidence/diagnostic-testing

[19] https://www.idsociety.org/practice-guideline/covid-19-guideline serology/#ExecutiveSummaryandBackground

Appendix 1: Preliminary Evidence to Decision

Table 1. Summary of initial judgements prior to the panel discussion (N=5/9)

FACTORS	JUDGEMENT						RESEARCH EVIDENCE/ADDITIONAL CONSIDERATIONS
Problem	No (N=1)	Yes (N=3)	Varies (N=1)				Yes, there are still institutions locally that make use of antibody testing in terms of crucial decision making for diagnosis and prior exposure to COVID-19
Benefits	Large	Varies (N=1)	Trivial (N=1)	Uncertain (N=3)			No evidence found on the benefits of screening
Harms	Large	Moderate	Small (N-1)	Uncertain (N=3)	Varies (N=1)		No evidence found on the harms of screening
Balance of Benefits and Harms	Favors the use of Antibody tests	Probably favors the use of Antibody tests (N=3)	Probably favors no diagnostic (N=1)	Don't Know (N=1)			
Certainty of Evidence	High	Moderate (N=2)	Low (N=1)	Very low (N=2)	No evidence (N=1)		DIAGNOSIS No evidence found
Accuracy	Very Accurate	Accurate (N=1)	Inaccurate	Very Inaccurate	Varies (N=2)	Don't Know (N=1)	PAST INFECTION The overall certainty of evidence is very low after downgrading for serious risk of

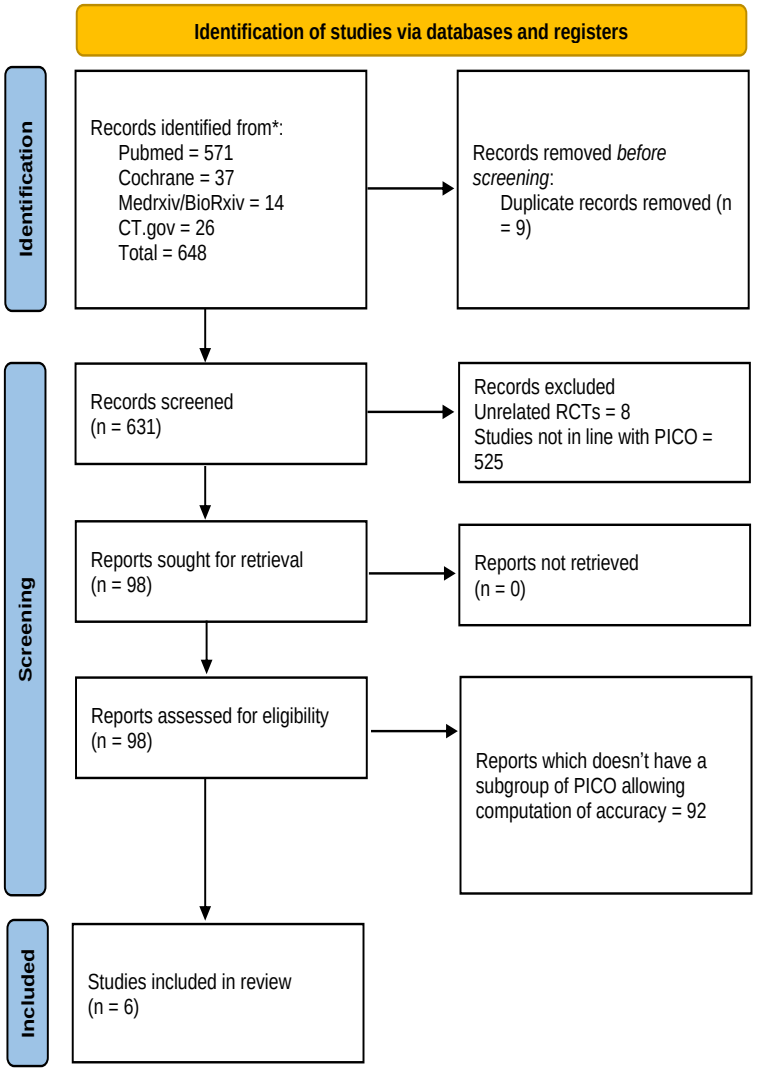
				(N=1)				bias and very serious inconsistency and very serious indirectness DIAGNOSIS No evidence found PAST INFECTION The pooled sensitivity of antibody tests was 99% (95% CI 96.7 – 99.7%; I2 = 73.2%; very low certainty) while pooled specificity was 11.9% (95% CI 2.5% - 41.4%; I2 = 73.2%; very low certainty) · Sensitivity was highest in the study that detected antibodies against the spike protein (n = 14,483): 100% (95% CI 100-100%) · specificity was highest in the study that detected antibodies against nucleocapsid protein (n = 3,708): 90% (95% CI 89-91%; very low certainty)
Values	Important uncertainty or variability (N=1)		Possibly important uncertainty or variability (N=3)	Probably NO important uncertainty or variability (N=1)	No important uncertainty or variability			No evidence found
Resources Required	Don't Know		Large cost (N=2)	Moderate Cost (N=2)	Negligible cost	Moderate savings	Varies (N=1)	Rapid antibody tests for COVID-19 may cost around ₱1500 to 2500

Certainty of evidence of required resources	No included studies (N=5)	Very low	Low	Moderate (N=1)	High		No local cost-effectiveness studies were found.
Cost effectiveness	No included studies (N=2)	Probably favors the comparator (N=1)	Does not favor either Antibody tests or the comparator (N=1)	Probably favors Antibody tests	Favors criteria		
Equity	Reduced	Probably Reduced (N=1)	Probably no impact (N=3)	Probably Increased (N=1)	Increased	Varies	
Acceptability	Don't Know	No	Probably No (N=3)	Yes (N=1)	Probably yes (N=1)	Varies	
Feasibility	Don't Know	No (N=1)	Probably No (N=1)	Yes (N=1)	Probably yes (N=2)	Varies	

Appendix 2: Search Yield and Results

Database	Search Terms	Date and Time	Yield
PubMed	(((COVID-19 OR SARS-COV-2) AND (serology OR serologic test OR antibody OR antibodies OR Immunoglobulin OR IgG OR IgM OR IgA)) AND (diagnosis OR accuracy OR sensitivity OR specificity OR predictive value)) AND (post vaccination OR vaccinated) Filter: From March 2021 to March 31 2023 Filter: Meta-analysis AND Review AND Systematic review AND Observational Studies	March 27, 2023	548
	(((seroprevalence[MeSH Terms]) AND (antibody OR serology[MeSH Terms])) AND (SARS-COV-2 OR COVID-19[MeSH Terms])) AND (Vaccinated OR post vaccination[MeSH Terms]) Filter: From March 2021 to March 31 2023	March 31, 2023	571
The Cochrane Library	COVID-19 OR SARS-COV-2 OR corona virus in Title Abstract Keyword AND Diagnosis OR diagnostic accuracy OR sensitivity OR specificity OR predictive value in Title Abstract Keyword AND Serology OR serologic test OR antibody OR immunoglobulin OR IgG OR IgA OR IgM in Title Abstract Keyword AND vaccinated OR post vaccination in Title Abstract Keyword Filter: From March 2021 to March 31 2023	March 27, 2023	4 reviews 358 trials
	COVID-19 OR SARS-COV-2 OR corona virus in Title Abstract Keyword AND serosurveillance OR seroprevalence in Title Abstract Keyword Filter: From March 2021 to March 31 2023	March 31, 2023	1 review 36 trials
MedRxiv & BioRxiv	abstract or title "COVID-19 OR SARS-COV-2 AND Serology OR serologic test OR antibody OR Ig"	March 27, 2023	2
	"Seroprevalence OR serosurveillance AND SARS-COV-2 OR COVID-19" Filter: From March 1 2021 to March 31, 2023	March 31, 2023	14
ClinicalTrials.gov	antibody test OR serology OR Serologic test OR IgG OR IgM OR IgA | SARS-COV-2 OR COVID-19 | accuracy OR sensitivity OR specificity OR predictive value	March 27, 2023	100
	SARS-COV-2 OR COVID-19 | Seroprevalence OR serosurveillance | Start date Filter: 03/01/2021 to 03/31/2023	March 31, 2023	26

Appendix 3: PRISMA Flow diagram for Serosurveillance studies



Appendix 4: Characteristics of Included Studies

Study ID	Setting	Index Test	Index Test Specimen	Population	Sample Size	Reference standard	Reference Standard Specimen
Blaszczuk 2022	Poland	Microblot-Array COVID-19 IgG assay (TestLine Clinical Diagnostics, Brno, Czech Republic) • Detects presence or absence of a reaction against at least 1 antigen—**NCP, RBD, or S2**	Serum	Medical staff in clinic hospital in Lublin Age: 20 – 65 years old (34.5% - 20 to 39 years old) Time of Ab testing: Between November 2021 to December 2021 Vaccine Type: 2 doses of Pfizer vaccine Timing of antibody testing: not specified	203 Subgoup of desired PICO = 168	Documented RT – PCR	Serum
murhekar 2021	India	IgG antibodies against **S1-RBD** (ADVIA Centaur XP/XPT, Siemens Healthineers, Munich, Germany) and **nucleocapsid** protein (Abbott ARCHITECT, Abbott Laboratories, Abbott Park, IL, US) of SARS-CoV-2 using chemiluminescence immunoassay	Serum	General Population and Healthcare workers in 70 Indian districts Age: 6 - >60 years old (70% >18 years old) Time of Ab testing: Between June to July 2021	36,227 Subgoup of desired PICO = 4,571	Reported Previous RT-PCR test	Serum

				Vaccine type: 1 to 2 doses of Covaxin (BBV152) or Covishield (ChAdOx1 nCoV-19) Timing of antibody test: atleast 21 days from first dose or 7 days from 2nd dose			
Cerbino-neto 2022	Brazil	**For adults:** SARSCoV-2 **Anti-S** IgG (Architect II, Abbott—Chicago, Illinois, EUA). • automated chemiluminescent microparticle immunoassay (CMIA) **For children and adolescent**: point-of-care (POC) lateral-flow rapid test (Fastep, Azure Tech. Co., Ltd.—Hangzhou, Zhejiang, China) • for **anti-N and anti-S1** IgG and IgM detection	Serum	Residents of Paqueta Island Age 0 - >60 years old (59% >60years old) Time of Ab testing: Between June 16-19 2021 Vaccine type: 1-2 doses – Astra Zenica vaccine 2 doses – CoronaVac 1 dose – Pfizer Timing of antibody test: 7 to 60 days from last vaccine dose	2,919 Subgoup of desired PICO = 954	Lab Confirmed, self-reported COVID-19	Serum
Decarreaux 2022	France	Euroimmun's semiquantitative anti-SARSCoV-2 (IgG) ELISA (reference: EI 2606-9601 G;	Serum	Staff and students of the University of Corsica, Corte, France	295	Lab confirmed (mixture of RT-PCR confirmed and	Serum

		EUROIMMUN, Bussy-Saint-Martin, France**)** • IgG antibodies to the SARS-CoV-2 **receptor binding domain (RBD) of the Spike protein** were evaluated		Age 18 to 64 years old (median age 37) Time of Ab testing: Between June 2021 to July 2021 Vaccine type: unspecified type, 1-2 doses Timing of antibody test: not specified	Subgroup of desired PICO = 186	Antibody (anti-S confirmed)	
Kanamori 2023	Japan	Elecsys Anti-SARS-CoV-2 (Roche Diagnostics) immunoassay was used with the Cobas e801 analyzer • detect **N-specific** total antibodies	Serum	HCWs in Juntendo University Hospital (JUH) who participated in annual health checkups in 2022 Age: 20 to >60 years old (31% - 30-39 years old, median 36 years old) Time of Ab testing: Between July to August 2022 Vaccine type: BNT162b2 and mRNA-1273 1-4 doses (89.3% - 3 doses)	3,788 Subgroup of desired PICO = 3,708	RT-PCR confirmed infection	Serum

				Timing of antibody test: not specified			
Thon 2023	Czech Republic	LIAISON SARS-CoV-2 S1/S2 IgG (DiaSorin, Saluggia, Italy) and SARS-CoV-2 IgG II Quant (Abbott, Sligo, Ireland) • IgG antibodies against the **spike protein**	Serum	Adult volunteers registered in the second largest health insurance company in Czech Republic Age: 18 to > 60 years old (30.28% - 40 to 49 years old) Time of Ab testing: Between April to Septmber 2021 Vaccine Type: Doses unspecified Mix of 1. Comirnaty (BioNTech Manufacturing, Mainz, Germany) 2. Spikevax (previously COVID-19 Vaccine Moderna; Moderna Biotech Spain, Madrid, Spain) 3. Vaxzevria (previously COVID-19 Vaccine AstraZeneca; AstraZeneca, Södertälje, Sweeden) 4. Jcovden (previously COVID-19 Vaccine Janssen; JanssenCilag International, Beerse, Belgium)	22,130 Subgroup of desired PICO = 14,483	Self-reported RT-PCR test	Serum

				Timing of antibody test: 5-7 months from vaccination			

Appendix 5: GRADE Evidence Profile

Question: Should Antibody testing be used to determien prior COVID-19 exposure in Vaccinated individuals?

Sensitivity	0.99 (95% CI: 0.97 to 1.00)
Specificity	0.12 (95% CI: 0.03 to 0.41)

Prevalences	10%	50%	90%

Outcome	№ of studies (№ of patients)	Study design	Factors that may decrease certainty of evidence					Effect per 1,000 patients tested			Test accuracy CoE	Importance
			Risk of bias	Indirectness	Inconsistency	Imprecision	Publication bias	pre-test probability of10%	pre-test probability of50%	pre-test probability of90%		
True positives (patients with COVID-19 disease)	6 studies 8759 patients	cross-sectional (cohort type accuracy study)	serious[a]	very serious[b]	very serious[c]	not serious	none	99 (97 to 100)	495 (484 to 499)	891 (870 to 897)	⨁◯◯◯ Very low	CRITICAL
False negatives (patients incorrectly classified as not having COVID-19 disease)								1 (0 to 3)	5 (1 to 16)	9 (3 to 30)		CRITICAL
True negatives (patients without COVID-19 disease)	6 studies 15311 patients	cross-sectional (cohort type accuracy study)	serious[a]	very serious[b]	very serious[c]	not serious	none	107 (23 to 373)	60 (13 to 207)	12 (2 to 41)	⨁◯◯◯ Very low	CRITICAL
False positives (patients incorrectly classified as having COVID-19 disease)								793 (527 to 877)	440 (293 to 487)	88 (59 to 98)		CRITICAL

Outcome	№ of studies (№ of patients)	Study design	Factors that may decrease certainty of evidence					Effect per 1,000 patients tested			Test accuracy CoE	Importance
			Risk of bias	Indirectness	Inconsistency	Imprecision	Publication bias	pre-test probability of10%	pre-test probability of50%	pre-test probability of90%		
Inconclusive	0 studies patients	-	-	-	-	-	-				-	
Complications	0 studies patients										-	

Explanations

a. Vulnerable to recall bias (3 studies use self-reported RT-PCR result and 1 study is a mixture of antibody test and RT-PCR result as a reference standard

b. studies included are all seroprevalence studies with just a subgroup of the desired PICO

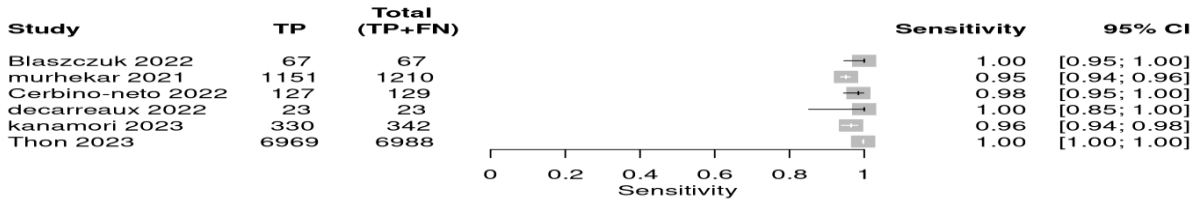
c. Substantial heterogeneity across studies (I^2 > 90%)

Appendix 5: Forest Plots

POOLED STUDIES

All studies

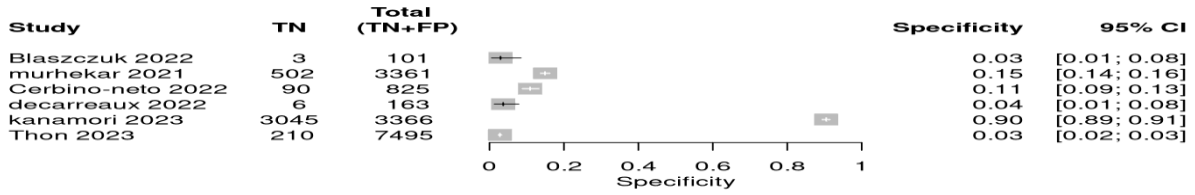
Heterogeneity: I^2 = 0.732

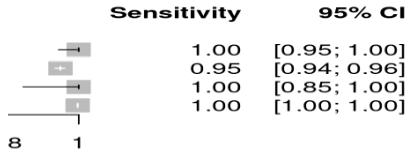


Heterogeneity: I^2 = 0.732

Antibody detected

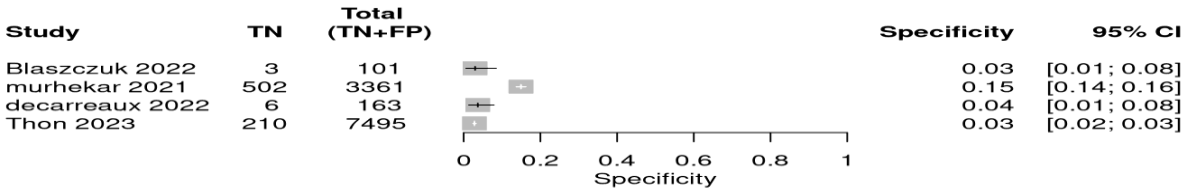
A. Studies using IgG antibodies





Heterogeneity: I^2 = 0.967

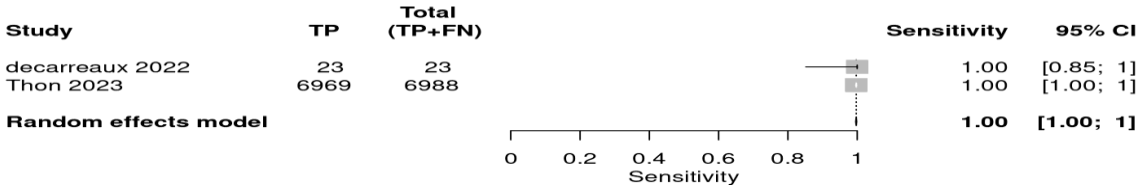
Heterogeneity: I^2 = 0.991

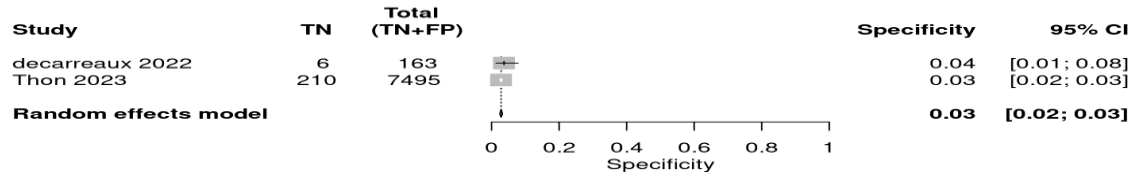


Specific antibody measured

A. Studies against spike protein

Heterogeneity: $I^2 = 0$

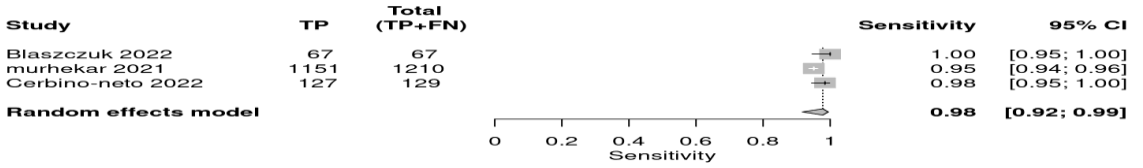




Heterogeneity: $I^2 = 0$

B. Studies against spike and nucleocapsid protein

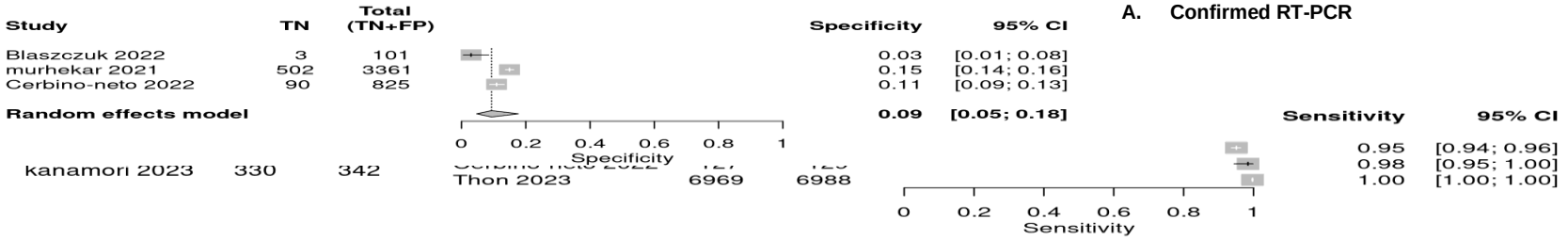
Heterogeneity: $I^2 = 0.245$



Heterogeneity: $I^2 = 0.883$

Reference standard

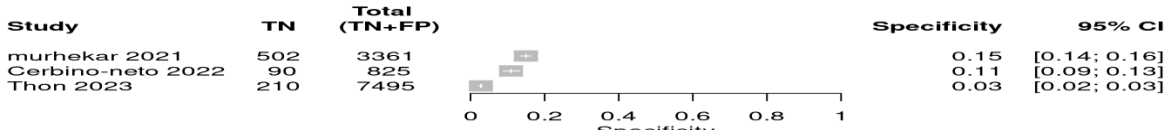
A. Confirmed RT-PCR



Heterogeneity: $I^2 = 0.0$

Antibody Tests

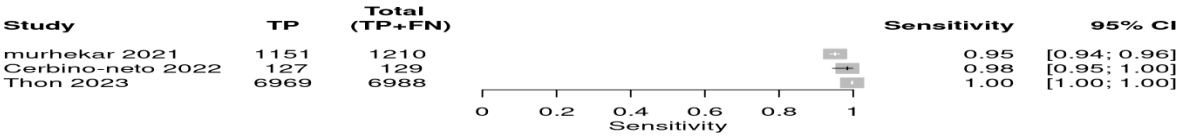
Heterogeneity: I^2 = 0.989



B. Self-reported RT-PCR

Heterogeneity: I^2 = 0.984

Heterogeneity: I^2 =



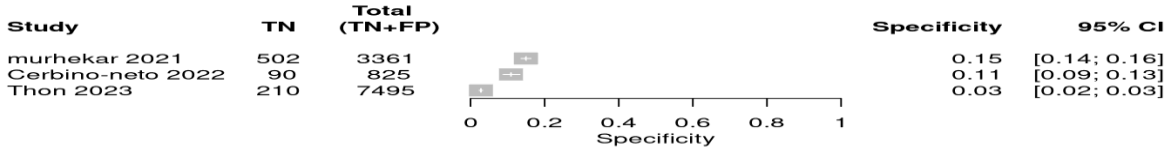
0.996

Population

A. General Population

Heterogeneity: I^2 = 0.984

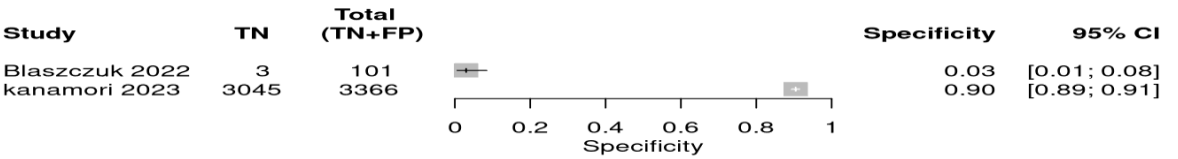
Heterogeneity: I^2 = 0.996



B. Healthcare workers

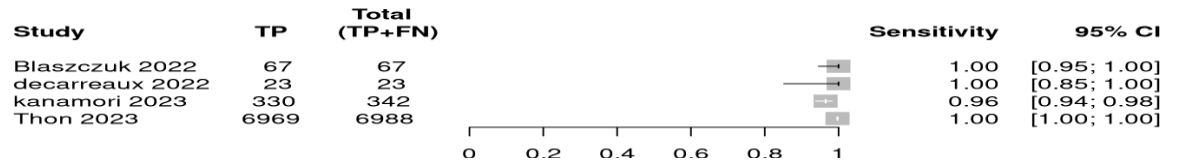
Heterogeneity: I^2 = 0.0

Heterogeneity: I^2 = 0.989



Antibody Tests

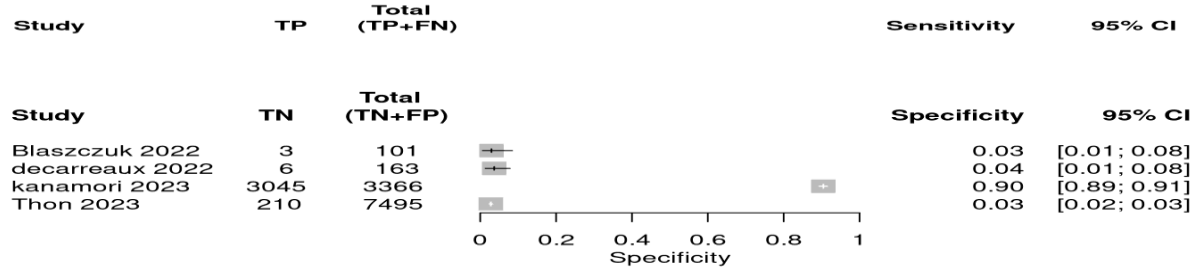
C. Adult



Heterogeneity: I^2 = 0.938

Heterogeneity: I^2 = 0.999

D. Adult and children



Heterogeneity: I^2 = 0.623

Heterogeneity: I^2 = 0.886

SINGLE STUDIES

1. Subgroups for Antibodies detected with single studies (Sp and Sn)

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)
Kanamori 2023	330	321	12	3045	0.96 [0.94, 0.98]	0.90 [0.89, 0.91]	0 0.2 0.4 0.6 0.8 1	0 0.2 0.4 0.6 0.8 1

A. Total antibodies

B. IgG and IgM

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)
Cerbino-neto 2022	127	735	2	90	0.98 [0.95, 1.00]	0.11 [0.09, 0.13]	0 0.2 0.4 0.6 0.8 1	0 0.2 0.4 0.6 0.8 1

2. Subgroups for specific antibody measured with single studies (Sp and Sn)

A. Nucleocapsid

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)
Kanamori 2023	330	321	12	3045	0.96 [0.94, 0.98]	0.90 [0.89, 0.91]	0 0.2 0.4 0.6 0.8 1	0 0.2 0.4 0.6 0.8 1

3. Subgroup for reference standard with single studies (Sp and Sn)

A. Antibody and RT-PCR

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)
Decarreaux 2022	23	157	0	6	1.00 [0.85, 1.00]	0.04 [0.01, 0.08]	0 0.2 0.4 0.6 0.8 1	0 0.2 0.4 0.6 0.8 1

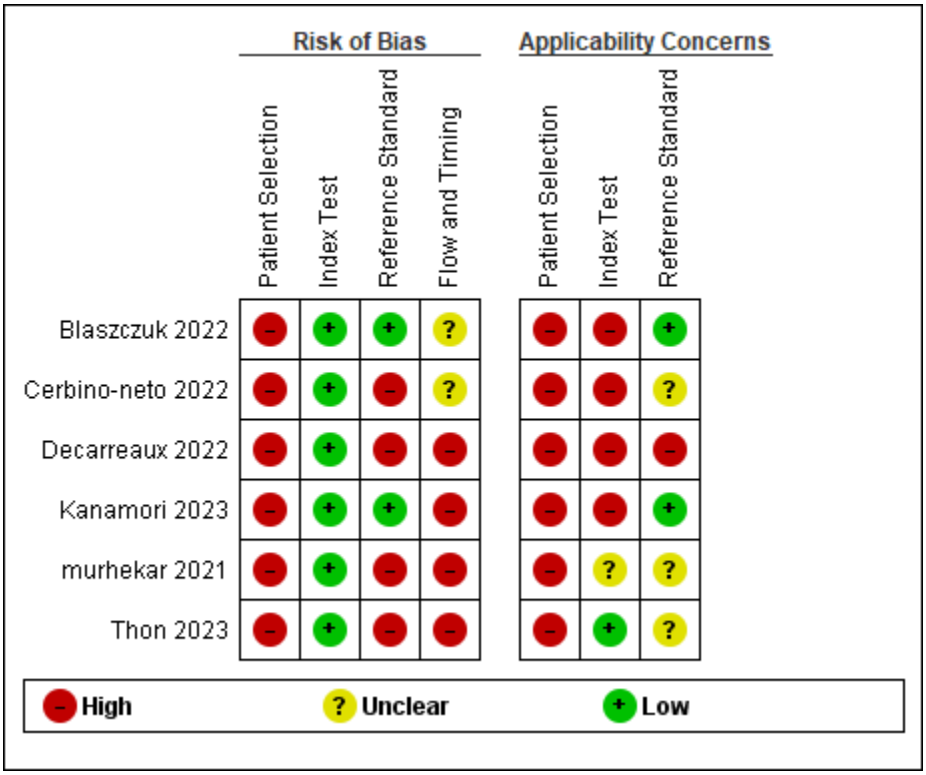
4. Subgroup for Population with single studies (Sp and Sn)

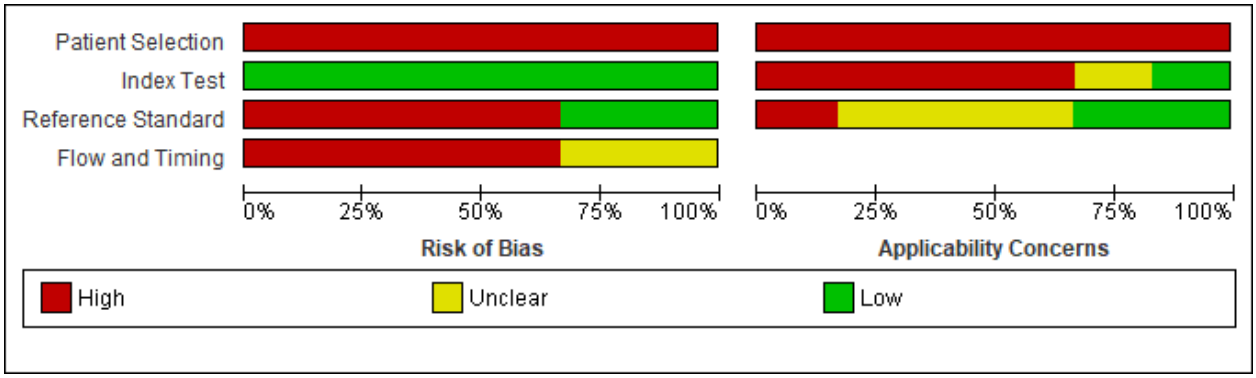
A. Healthcare worker + General Population

Study	TP	FP	FN	TN	Sensitivity (95% CI)	Specificity (95% CI)	Sensitivity (95% CI)	Specificity (95% CI)
murhekar 2021	1151	2859	59	502	0.95 [0.94, 0.96]	0.15 [0.14, 0.16]	0 0.2 0.4 0.6 0.8 1	0 0.2 0.4 0.6 0.8 1

Appendix 6: Study Appraisal

Risk of bias and applicability concerns graph: review authors' judgements about each domain presented as percentages across included studies





Appendix 7: Subgroup Analyses

Subgroup	No. of studies	Sensitivity (95% CI)	I^2	Specificity (95% CI)	I^2
ALL STUDIES	6	0.99 (0.967 – 0.997)	0.732	0.119 (0.025 – 0.414)	0.732
Population					
Healthcare workers	2	0.97 (0.95 – 0.98)	0%	0.35 (0.01 – 0.97)	98.9%
General Population	3	1.00 (1.00 – 1)	98.4%	0.05 (0.02 – 0.10)	99.6%
HCW and Gen. Pop	1	0.95 (0.94 – 0.96)	-	0.15 (0.14 – 0.16)	-
Adult	4	0.99 (0.97 – 1.0)	93.8%	0.12 (0.01 – 0.61)	99.9%
Adult + children	2	0.95 (0.94 – 0.96)	62.3%	0.13 (0.10 – 0.16)	88.6%
Specific antibody measured					
Spike Protein	2	0.997 (0.996 – 0.998)	0%	0.028 (0.025 – 0.032)	0%
Nucleocapsid	1	0.96 (0.94 – 0.98)	-	0.90 (0.89 – 0.91)	-
Both N + S	3	0.978 (0.921 – .994)	24.5%	0.094 (0.048 – 0.176)	88.3%
Antibody Detected					
IgG only	4	0.99 (0.96 – 1.00)	96.7%	0.05 (0.02 – 0.10)	99.1%
Total antibody	1	0.96 (0.94 – 0.96)	-	0.90 (0.89 – 0.91)	-
IgG and IgM	1	0.98 (0.96 – 1.0)	-	0.11 (0.09 – 0.13)	-
Reference standard					
Self-reported RT-PCR	3	0.99 (0.95 – 1.00)	98.4%	0.08 (0.03 – 0.17)	99.6%
Confirmed RT-PCR	2	0.97 (0.95 – 0.98)	0%	0.35 (0.01 – 0.97)	98.9%
Ab and RT-PCR	1	1.00 (0.85 – 1.0)	-	0.04 (0.01 – 0.08)	-