Philippine COVID-19 Living Clinical Practice Guidelines
Institute of Clinical Epidemiology, National Institutes of Health, UP Manila
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EVIDENCE SUMMARY

Among COVID-19 confirmed patients, should certain RT-PCR cycle threshold values be used to determine infectivity?

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RECOMMENDATION

There is insufficient evidence to recommend an RT-PCR cycle threshold cut-off value to determine infectivity among COVID-19 confirmed patients. Interpretation of RT-PCR cycle threshold values may vary and is dependent on the PCR assay used, gene target, sample type, and timing of sample collection. *(Very low certainty of evidence)*

Consensus Issues
The panel recognizes that RT-PCR cycle threshold values may have utility specifically for patients with previously documented COVID-19 infection. However, evidence remains insufficient on the cut-off cycle threshold value that differentiates infectious virus from viral remnants.

Key Findings

- Twelve observational studies were included in this review on the use of cycle threshold (Ct) as a surrogate marker of infectivity as evidenced by viral isolation in culture. One systematic review and meta-analysis was included on the association of Ct value with patient clinical outcomes.
- Very low certainty evidence showed that among COVID-19 cases, Ct values of 24 to 35.6 were associated with isolation of SARS-CoV-2 virus in culture. Lower Ct values (Ct < 25-30) were associated with increased disease severity and mortality.
- Among convalescent and clinically recovered patients with persistent positive RT-PCR test, Ct values ranged from 30 to 41.7. Samples from these cases had a low yield of virus isolation in culture and had degraded viral fragments on genome sequencing.
- Interpretation of Ct values must be done with caution due to variations in PCR assay methods, target gene, sample type, and timing of sample collection.

Introduction

Cycle threshold (Ct) value is defined as the number of cycles of amplification required for the fluorescence of a PCR product to exceed the background signal and be detected to cross a threshold of positivity. Ct values are inversely related to viral load and can provide an indirect method of quantifying the copy number of viral RNA in a sample. Because of its correlation with viral load, some studies have suggested that lower Ct values may be associated with worse outcomes and may carry some prognostic value.[1] However, Ct values cannot be directly compared across assays, and must be interpreted with caution as these are influenced by sample type, timing of sample collection, assay design, and pre-analytic issues. It is currently unclear whether certain Ct values could be used as a marker of infectivity and as a guide to patient management decisions.[2-4]

Review Methods

Comprehensive literature search in MEDLINE, Cochrane Library, and Scopus was done last September 23, 2021 using search terms related to SARS-CoV-2, COVID, cycle threshold, viral

load, and viral culture. Studies were included if these (a) involved patients with suspected or confirmed COVID-19 tested using RT-PCR, (b) used upper or lower respiratory tract samples (e.g., oropharyngeal, nasopharyngeal, sputum, endotracheal aspirate) for RT-PCR, and (c) confirmed infectivity of these samples through viral isolation in culture. Study designs were limited to observational, cross-sectional studies, or systematic reviews of observational studies. The detailed search strategy is found in Appendix 2.

Methodological quality of the included observational studies was appraised using the Newcastle Ottawa Scale for cross-sectional studies.[5,6] When possible, subgroup analysis was performed to explore the influence of covariates (e.g., severity, symptom status, target gene) on the Ct values. Methodological quality of the systematic review and meta-analysis was appraised using the Painless Evidence Based Medicine critical appraisal of systematic reviews and meta-analyses.[7]

Results

Summary of characteristics of included studies

The aforementioned search strategy yielded a total of 829 studies, which included four systematic reviews.[1,7-9] After screening for eligibility, removal of duplicates, and cross-referencing, 12 studies [10-21] and one systematic review and meta-analysis [22] were included in this review. Nine cross-sectional, one longitudinal, one cohort studies, and one brief report were done in various locations in Europe (United Kingdom, France, Austria, Italy, Spain), North America (Canada, United States of America), Australia, and Asia (Taiwan, Singapore). The studies were all peer reviewed except for two studies, among which one was a pre-proof and the other a brief report.[11,15]

The 12 observational studies had a total of 1,728 respiratory tract samples (oropharyngeal, nasal, and nasopharyngeal swabs, sputum, and bronchial aspirates) from COVID-19 confirmed subjects (active and recovered cases, symptomatic and asymptomatic cases, varying disease severities, healthcare workers, inpatients and outpatients, and nursing home residents and staff) that were analyzed. Duration of data collection ranged from one to five months where the earliest sampling was done in January 2020 and latest sampling last August 2020. Timing of sample collection relative to symptom onset ranged from 0 to 51 days. Ct values from various target genes (E, N, M, nsp12, S, nsp 2, RdRp, ORF1ab) corresponding to samples with viral isolation in culture were identified. Specific PCR assays used varied across studies and were not identified in some studies.

With regard to the association between Ct values and other clinical outcomes (e.g., hospitalization, disease severity, and mortality), data were obtained from a systematic review [22] that included 7 studies (n=3,291).

The characteristics of the included studies is detailed in Appendix 3.

Summary of results of included studies

A. Ct value and clinical outcomes

Seven studies from the systematic review and meta-analysis by Shah et al. [22] revealed no difference in the mean Ct values for those who were hospitalized when compared to those who were not hospitalized (MD 0.062, 95% CI -1.933 to 2.056). There was a statistically significant increase in disease severity (OR 2.31 95% CI 1.7-3.13) and increase in mortality (OR 2.95, 95% CI 2.19-3.96) of lower Ct values (<25 or 25-30) when compared with higher Ct values (>30).

B. Ct value and viral isolation in culture (infectivity)

Eleven out of 12 studies yielded isolation of virus in culture. Ct value corresponding to viral isolation in culture ranged from 24 to 35.6. Singanayagam et al. [9] estimated that the odds

ratio of recovering infectious virus decreased by 0.67 for each unit increase in Ct value (95% CI 0.57-0.77), while Bullard et al. [16] demonstrated an odds ratio for positive viral culture of 0.64 (95% CI 0.49-0.84) for every 1 unit increase in Ct. However, these studies had varied population studied, PCR assay used, target gene investigated, and culture methods and techniques employed.

Table 1. Cycle threshold cut off value associated with viral isolation in culture

Study	Specimen	Target gene	Cut off Ct value associated with viral isolation
Bullard 2020 [16]	NP, ETT	E	24
Brown 2020 [15]	Nasal, throat swab	RdRp, E, N, ORF1lab	26.2
Young 2020 [20]	NP	Not stated	30
Basile 2020 [18]	Mixed URT + LRT	E, RdRp, N, M, ORF1 lab	32
Gniazdowsky 2020 [17]	NP	S, Nsp2	32.1
La Scola 2020 [10]	NP, sputum	E	33
Folguiera 2021 [14]	NP, bronchial aspirates	E	35
Ladhani 2020 [13]	Nasal swab	ORF1lab	35
Singanayagam 2020 [9]	Mixed URT	RdRp	35
Huang 2020 [19]	NP, OP, sputum	E, N, nsp12	35.2
Piralla 2020 [12]	Nasal	E, N	35.6

ETT: endotracheal tube aspirate, LRT: lower respiratory tract, NP: nasopharyngeal, OP: oropharyngeal, URT: upper respiratory tract.

C. Subgroup Analysis

Symptomatic vs asymptomatic

One study revealed no statistically significant difference in the Ct value and live virus recovery of symptomatic and asymptomatic cases.[10] In this study by Singanayagam et al., the reported median Ct was 31.23 (IQR 28.21-32.97) in asymptomatic cases, 30.94 (IQR 27.08-34.57) in mild to moderate cases, and 32.55 (IQR 28.39-33.66) in severe.[10] Culture positivity rates were also comparable in symptomatic and asymptomatic cases, with an estimated OR of 0.66 (95% CI 0.34-1.310).[10]

Clinically recovered or convalescent cases

Two studies included patients who were clinically recovered or labelled as 'convalescent' with either persistently positive SARS-CoV-2 RT-PCR or had a positive result after a negative result. The timing of sample collection had a median of 23 days (range 5-51 days) from the day of RT-PCR positivity or a median of 37 days (range 19-58 days) from symptom onset [11] or during their time of discharge or continuation of isolation period.[12] Laferl et al. [12] recorded a mean Ct value of 37.4, median of 37.3, and range of 30.8 to 41.7. No virus was isolated in all the samples collected. Piralla et al. [12] reported the viral load in clinically recovered patients at the time of discharge with a median Ct value of 36.8 (range 30-39.4). Furthermore, only 2.3% of these samples yielded viral isolation in culture and subsequent genome sequencing of these samples revealed non-functional viral genomes. Table 2 lists the studies on clinically recovered and convalescent cases.

Table 2. Characteristics of studies on clinically recovered and convalescent cases

Study	Population	PCR Assay	Timing of sample collection	Target Gene	Sample Type	Ct value	Culture Positivity	Genome Sequencing
Laferl et al. [12]	Convalescent HCW	Thermo Fisher Scientific USA	23 days (range 5-51 days) from day of RT PCR positivity or median of 37 days (range 19-58 days) from symptom onset	E	OPS, NPS	**Mean**: 37.4 **Median**: 37.3 **Range**: 30.8 -41.7	**0%**	Not done
Piralla et al. [13]	Clinically recovered COVID-19 patients	Seegene Allplex 2019 nCOV assay	During the time of discharge or during isolation	E, N	Nasal swab	**Median**: 36.8 **Range**: 30-39.4	**2.3%** **Median Ct value (culture positive):** 35.6 **Median Ct value (culture negative):** 36.9 P = 0.37	Samples from positive culture did not have the whole viral genome ~ non functional residual RNA

Target gene

Huang et al. [20] compared the Ct values of culture-positive and culture-negative from three target genes: N, E, and nsp12. For the 3 genes, Ct values from culture-positive samples were lower than culture-negative samples. N as target gene showed higher Ct values compared to nsp12 and E as target genes for the same sample.

Table 3. Ct values of culture-positive and culture-negative samples per target gene

Genes	Culture Positive		Culture Negative	
	Mean Ct value (SEM)	Range	Mean Ct value (SEM)	Range
Nsp 12	23.9 (0.78)	17.75-31.47	29.26(0.69)	22.32-36.52
E	22.39 (0.75)	16.85-31.46	28.92 (0.65)	20.89-38.33
N	27.29 (0.77)	22.14-35.2	27.29(0.77)	22.14-35.2

Type of sample

Basile et al. revealed a statistically significant difference in the Ct values of upper respiratory tract samples and lower respiratory tract samples of active inpatient and outpatient cases.[19] Higher Ct values were recorded from samples of the upper respiratory tract (mean Ct 26.76) as compared to the lower respiratory tract (mean Ct 34.41).

Whole Genome Sequencing

Two studies performed whole genome sequencing in samples that yielded virus isolation in culture.[13,20] In the samples coming from 50 active cases [20], culturable specimens had higher correlation between structural and non-structural genes. Non-culturable specimens were characterized by higher or lower nsp12 RNA level, suggesting existing degraded

intermediates. In samples of clinically-recovered cases who persistently tested positive for RT-PCR [13], genome sequencing revealed non-functional and residual RNA.

Ongoing Studies
No ongoing studies were found in the search strategy done.

Certainty of evidence

For the association of Ct values with viral isolation in culture, the overall certainty of evidence was rated very low due to serious risk of bias and inconsistency. All studies were assessed to have unclear to high risk for selection bias due to either unclear sampling method or use of convenience sampling. Majority of the included studies lack statistical methods for justifying sample size and controlling confounders. Serious inconsistency noted was related to the heterogeneity in RT-PCR characteristics and Ct values reported.

For the association of Ct values with clinical outcomes, the overall certainty of evidence was rated very low due to moderate to high risk of bias and heterogeneity brought about by difference in assay characteristics.

The table of risk of bias scoring is shown in Appendix 4.

Other Considerations

Table 4 lists other considerations on the utility of Ct values.

Table 4. Other considerations on the utility of Ct values.

Equity	Any patient subjected to RT-PCR testing can retrieve its Ct value with no additional cost.
Cost	RT-PCR (DOH Price Cap): ● Minimum P3,800.00 ● Maximum P4,500.00
Acceptability	Any patient subjected to RT PCR testing can retrieve its cycle threshold value with no additional processing and analysis required.
Feasibility	Any patient subjected to RT PCR testing can retrieve its cycle threshold value with no additional processing and analysis required. Ct value determination is part of the routine processing of RT-PCR, but is not routinely reported in the official results. Cut off values are machine dependent.

Recommendations from Other Groups

The **United States Center for Disease Control** (CDC) released information on the utility of Ct values in their Frequently Asked Questions [3] last August 25, 2021. The response stated that Ct values from different RT-PCR tests cannot be compared due to differences in nucleic acid target, platform, etc. Ct values should not be used to determine an individual's viral load, level of infectiousness, or eligibility to be released from isolation or quarantine. From a public health perspective, however, CDC maintains that median Ct values from a population or group may be valuable for public health to evaluate viral load and transmissibility for a particular SARS-CoV-2 variant or to compare the viral load between two groups (e.g., vaccinated versus unvaccinated individuals).

Public Health England [2] released a guide for health protection teams which stated that Ct values are not directly comparable between assays and may not be reported by some RT-PCR platforms in use. Interpreting single positive Ct values for staging infectious course,

prognosis, infectivity, or as indicator of recovery must be done with context about the clinical history.

On the other hand, **Public Health Ontario** [23] stated that SARS-CoV-2 Ct values may be of use when interpreting positive laboratory results derived from patients with low pretest probability, in particular, asymptomatic persons with no epidemiologic link to a confirmed COVID-19 case.

Research Gaps

Additional studies are required to investigate the utility of Ct values and cell culture results in making clinical decisions and developing diagnostic strategies that can differentiate shedding versus active replication and will be very valuable for infection control.[12] Larger studies are needed to establish Ct criteria that reliably correlates with loss of infectivity and that utilize additional gene targets.

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Appendix 1. Evidence to Decision

Table 1. Summary of initial judgements prior to the panel discussion (N = 7)

FACTORS	JUDGEMENT					RESEARCH EVIDENCE/ADDITIONAL CONSIDERATIONS
Problem	No	Yes (7)				Surrogate marker for infectiousness is important in patient management decisions since the gold standard, viral culture, is costly and would need more resources and time.
Benefits	Large (1)	Moderate (3)	Small (3)	Uncertain		Cycle threshold may have the potential to be a surrogate marker of infectiousness. However, no effect estimates can be generated from the evidences reviewed on the benefit of cycle threshold determination.
Harms	Large (1)	Moderate	Small (3)	Uncertain (3)		No evidence on the harm of cycle threshold determination.
Balance of Benefits and Harms	Favors the use of RT-PCR Ct values (1)	Probably favors the use of RT-PCR Ct values (3)	Does not favor the use of RT-PCR Ct values (3)			No evidence on the effect estimates generated on the benefit or harm of cycle threshold determination.
Certainty of Evidence	High (1)	Moderate	Low (1)	Very low (5)		The overall certainty of evidence was very low due to fair risk of overall bias attributed to selection bias and lack of statistical methods for sample size justification and control of confounders in some studies.
Accuracy	Very Accurate	Accurate (1)	Inaccurate (1)	Very Inaccurate	Uncertain (5)	No evidence on the diagnostic accuracy.
Values	Important uncertainty or variability	Possibly important uncertainty or variability (3)	Possibly NO important uncertainty or variability (3)	No important uncertainty or variability (1)		No evidence found.

FACTORS	JUDGEMENT						RESEARCH EVIDENCE/ADDITIONAL CONSIDERATIONS
Resources Required	Uncertain (1)	Large cost	Moderate Cost	Negligible cost (5)	Moderate savings	Large savings (1)	There is no added cost when cycle threshold is requested in addition to RT-PCR. RT-PCR minimum cost is P3,800.00, maximum cost or price cap at P4,500.00 (DOH).
Certainty of evidence of required resources	No included studies (3)	Very low (1)	Low (2)	Moderate (1)	High		Cost of RT-PCR may vary depending on the hospital or laboratory.
Cost effectiveness	No included studies (4)	Favors comparator	Does not favor either RT-PCR Ct values or the comparator (2)	Favors the use of RT-PCR Ct values (1)			No cost effectiveness study is available
Equity	Uncertain (3)	Reduced (2)	Probably no impact (1)	Increased (1)			RT-PCR is the standard test to confirm COVID-19 disease which has been made available to the public with reduced cost and can be subsidized by PhilHealth.
Acceptability	Uncertain (2)	No	Yes (5)				RT-PCR is the standard test to confirm COVID-19 disease which can provide Ct values. No additional procedure to be done to determine Ct on the part of the patient, doctor, and laboratory personnel apart from the usual OPS/NPS and laboratory processing of RT-PCR.
Feasibility	Uncertain	No	Yes (7)				Ct value determination is part of the RT-PCR process.

Appendix 2. Search Yield and Results

Data Base	Search Strategy	Yield
Medline	(Corona virus disease OR COVID-19 OR SARS CoV 2) AND (cycle threshold OR cycle threshold value OR Ct value) AND (Viral load) Filters: Free full text, Meta-analysis, Review, Systematic Review	328
Cochrane	(COVID 19 OR SARS CoV 2) AND (cycle threshold OR Viral load OR viral culture)	467
Scopus	(COVID 19 OR SARS CoV 2) AND (cycle threshold OR Viral load OR viral culture)	34

Appendix 3. Characteristics of Included Studies

Table 1. Included studies on the association of Ct values with viral isolation in culture

Study [Reference Number]	Country	Design	Peer reviewed	Number of samples (patients)	Population	Sample type	RT-PCR Target	Timing of RT-PCR testing	Outcome
Singanayagam 2020 [10]	United Kingdom	Cross-sectional, retrospective, single center (NRL)	Yes	324 (253)	Samples from COVID-19 patients from range of clinical scenarios (community, HCW surveillance, outbreak, asymptomatic close contacts) 92% - asymptomatic/mild-to-moderate cases	Mixed URT (some self sampled)	RdRp gene	Not reported	Median Ct values for asymptomatic, mild-to-moderate, severe cases Odds ratio (of recovering infectious virus for each unit increase in Ct value)
La Scola 2020 [11]	France	Cross Sectional, retrospective, single center	(Only Brief Report/ Preliminary Clinical Study)	183 (155)	Samples of COVID-19 patients received at the sole diagnostic center in Marseille	NPS 95% Sputum 5%	E gene	Not reported	RT-PCR Ct value and viral isolation in culture
Laferl 2020 [12]	Austria	Longitudinal, prospective, single center	Yes	58 (15) 24/58 RT-PCR positive	Convalescent healthy HCW with COVID-19 in isolation (13/15, symptomatic), Ct $\leq$ 42	OPS 50% NPS 50%	E gene	median 23 days (range 5-51 days RT-PCR positivity); median 37 days (range 19 – 58 days) from symptom onset	RT-PCR Ct value and viral isolation in culture IgM, IgG
Piralla 2020 [13]	Italy	Cross-sectional, multi-center, prospective	Yes	387 (number of patients not specified)	COVID-19 patients, clinically recovered with low viral load Ct > 30 (persistently positive) 'convalescent patients' HCW, hospitalized, part of epidemic response	nasal	E and N genes	Time of discharge or quarantine period	% positivity RT-PCR Ct value (positive vs negative culture) Genone Sequencing
Ladhani 2020 [14]	London	Single Population Cohort,	Yes	158 cases (105	COVID-19 lab confirmed residents and healthcare workers of nursing homes (Ct < 35)	Nasal swabs	ORF1ab gene	Not stated (status taken at Day 0, Day 14)	%positivity RT-PCR CT values

Study [Reference Number]	Country	Design	Peer reviewed	Number of samples (patients)	Population	Sample type	RT-PCR Target	Timing of RT-PCR testing	Outcome
		prospective, multicenter		residents, 53 staff)					Whole genone sequencing
Folgueira 2020 [15]	Spain	Cross-sectional, retrospective, single center	No	106 (105)	COVID-19 lab confirmed outpatient (Mild, HCWs) and hospitalized (severe) patients	102 nasopharyngeal exudates and 4 bronchial aspirates (ICU cases)	E gene	Mild cases median 3 days Severe median 6 days	% positivity RT-PCR CT value (mild vs severe forms)
Brown 2020 [16]	England	Cross Sectional, prospective, Multicenter	Yes	23 HCW	Healthcare workers	Nasal and throat swab	ORF1ab; RdRp, E, and N	Various presentation (asymptomatic, presymptomatic, symptomatic) 17 previous symptomatic median day of symptom 27 days (range 3-43 days)	% positivity RT-PCR CT values
Bullard 2020 [17]	Canada	Cross-sectional, retrospective	Yes	90 samples	Samples of COVID suspects	NP and ETT	E gene	Not stated	RT-PCR Ct value and viral isolation in culture STT
Gniazdowski 2020 [18]	USA	Cross-sectional, single center, retrospective	Yes	131 samples	COVID-19 patients hospitalized in JHH	NPS	S gene, Nsp2 gene	Not stated	Ct value and viral isolation in culture Prolonged positive Positive after negative WGS ddPCR
Basile 2020 [19]	Australia	Cross-sectional, retrospective	Yes	56 samples	COVID-19 patients with varying severity (91%-outpatient, 6.1%-inpatients, 2.6%-critical) 1. Routine lab as part of outbreak	Mixed URT and LRT	E, RdRp,N,M, ORF1ab (Ct at N gene)	0-29 days	Ct values at N gene and virus isolation in culture

Study [Reference Number]	Country	Design	Peer reviewed	Number of samples (patients)	Population	Sample type	RT-PCR Target	Timing of RT-PCR testing	Outcome
					2. ICU cases Persistent positive				
Huang 2020 [20]	Taiwan	Cross-sectional, retrospective	Yes	60 (50)	COVID-19 lab confirmed cases in virology lab	OP, NP, sputum	nsp12, E, and N genes	Not stated	%positivity RT-PCR Ct value (positive vs negative culture) Genome copy numbers
Young 2020 [21]	Singapore	Single Population Cohort, prospective, multi center	Yes	152 (74)	COVID-19 lab confirmed	Nasopharyngeal	Not stated	Various severities	%positivity RT-PCR CT values Disease severity Seroconversion

Table 2. Included studies on the association of Ct values with clinical outcomes

Study [Reference Number]	Country	Design	Peer Review	Number of Subjects	Population	Sample Type	Outcome
Shah et al [22]	Minnesota, USA	Systematic Review and Meta-analysis	Yes	13885 (18 studies) 3291 (7 studies for meta-analysis)	COVID-19 confirmed cases	NP or NP/OPS	Hospitalization Disease severity Mortality

Appendix 4. Study Appraisal

Table 1. Newcastle-Ottawa Scale for appraisal of cross-sectional studies

	Singanayagam [10]	La Scola [11]	Laferl [12]	Piralla [13]	Ladhani [14]	Folgueira [15]	Brown [16]	Bullard [17]	Gniazdowsky [18]	Basile [19]	Huang [20]	Young [21]
SELECTION												
Representativeness of the sample	0 Sampling design not stated	0 Sampling design not stated	0 Unclear, subjects by invitation	0 Sampling design not stated	0 Sampling design not stated	0 Sampling design not stated	0 Convenience sampling	0 Sampling design not stated	0 Sampling design not stated (hospitalized with comorbidities)	0 Sampling design not stated	0 Sampling design not stated	0 Sampling design not stated
Sample Size	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	1	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated
Non Respondents	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated	0 Not stated
Ascertainment of exposure	1	1	1	1	1	1	1	1	1	1	1	1
COMPARABILITY												
Comparable subjects in different outcome groups, based on study design or analysis, confounders controlled	1 Subgroup analysis done	0 Not stated	0 Not stated	1 Subgroup analysis done	1	0 Not stated	1 MVA done	1	0 Not stated	0 Not stated	1 Subgroup analysis done	0 Not stated
OUTCOME												
Assessment of outcome	1	1	1	1	1	1	1	1	1	1	1	1
Statistical test	1	1	Not stated	1	1	1	1	1	1	1	1	1
TOTAL	4	3	2	4	4	3	5	4	3	3	4	3

Table 2. Critical Appraisal of systematic review and meta-analysis (Shah et al.)

Criteria	Appraisal
1. Appraising directness	Direct P – COVID -19 cases E – Ct values O – Infectivity for guidance of patient management decisions*
2. Appraising validity (inclusion criteria, search strategy, validity assessed, reproducible)	Valid Inclusion criteria stated Comprehensive literature search done January 28, 2021 Moderate to high risk of bias 2 reviewers
3. Appraising the results (overall results, precision, heterogeneity)	Odds Ratio and MD reported Imprecision noted (wide CI), Significant heterogeneity (I2 > 50%)
4. Applicability	For CPG development
5. Individualizing results	For CPG development

* *In this systematic review, Ct value was used as a surrogate marker of viral load for prognostication which may guide patient management decisions*