

COVID-19 Scientific Advisory Group

Rapid Evidence Report

How do the characteristics for COVID-19 testing differ between samples collected from nasal, nasopharyngeal, and/or throat swabs compared to saliva samples?

1. Do the testing characteristics vary between nasal, nasopharyngeal, and/or throat swabs vs. saliva samples?
2. Does the sensitivity and specificity of SARS-CoV-2 non-serologic tests vary between asymptomatic or symptomatic patients?

Context

- In the initial stages of the COVID-19 response, [a review comparing the testing characteristics of samples retrieved from nasopharyngeal, nasal, and throat swabs](#) was conducted by the Scientific Advisory Group to address concerns of variability between the various sample collection methods
- Since April 2020, the body of evidence for COVID-19 testing has grown substantially as testing strategies have evolved. In addition, saliva is now recognized as a potential sample with which to test for COVID-19. It is unclear how saliva compares to conventional nasal, nasopharyngeal (NP), or throat/oropharyngeal (OP) swabs.
- In August 2020, a brief [review on the testing characteristics of self-collected samples](#) was conducted. Thus, the testing characteristics of samples that have been self-collected is out of scope for this update.
- There is some interest in using saliva as a specimen for testing in the community, rather than acute care. The relative ease of collection of saliva makes it a potentially attractive option for self-collection and submission for testing.

Key Messages from the Evidence Summary

- The body of evidence on testing models is of low-moderate quality. The body of evidence regarding saliva as a specimen is of reasonable quality, as is the small body of evidence regarding testing in asymptomatic individuals. It is important to note that the evidence on this topic is rapidly evolving and meta-analytic findings should be considered carefully rather than accepted as truth.
- A meta-analysis of NP, OP, and sputum samples from hospitalized patients found that sputum had the highest percentage of positive results (71%), followed by NP swabs (54%) and OP swabs (43%). In studies included here that compared multiple methods of specimen collections, nasopharyngeal and oropharyngeal swabs were often used interchangeably and together had a sensitivity ranging from 60% (5 days post symptom onset) to 97.8%. One study showed that nasal swabs had a sensitivity of 87%.
- Local data generated by Alberta Precision Laboratories (APL) demonstrate near equivalence of NP and OP swabs while nasal swabs performed suboptimally.
- “Saliva” is a poorly defined term in the literature and is notably inconsistent. The sample can be derived from several places in the oral cavity and by multiple methods, and the utility of the sample can be affected by the intensity of disease, time of day, activity (eg. smoking, eating, and drinking), collection method (eg. drooling, spitting, or swabbing), use of transport media and type if used, and storage environment. The evidence in the primary literature is mixed regarding the best collection method, however, the meta-analysis by Peeters (in preprint) suggests that there is no difference between spitting into vials and oral swabbing.

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- The testing characteristics of saliva are highly variable. The sensitivity of saliva was found to range from 31-100%, while specificity of saliva ranges from 71-100%. Pooled estimates from meta-analyses show sensitivity ranges from 83.4% - 97%; a pooled estimate of specificity suggests 97.7%.
- Generally, saliva appears to be a less sensitive specimen type than a NP or OP swab. Under ideal specimen collection and processing conditions that may not be achievable in Alberta, saliva may be comparable to conventional swabbing methods as a specimen.
- The body of literature on the testing characteristics of specimens from asymptomatic vs. symptomatic individuals is small and in this review, opportunistic. No studies were identified in the database search that directly compared asymptomatic to symptomatic individuals. In studies where the sampling method was biased towards symptomatic and confirmed COVID-19 cases (thus artificially inflating disease prevalence), it appears that testing samples from asymptomatic individuals is less sensitive than testing samples from symptomatic individuals. Conversely, one large study of asymptomatic testing with a low disease prevalence (2.9%) suggests that the sensitivity of specimens from asymptomatic individuals is high (90%), with concordance probability ranging from 0.93-0.99 as prevalence was tested from 0-30% (Yokota et al., 2020).

Committee Discussion

The committee agreed with the recommendations as presented here and held a robust discussion about the role that saliva might play in Alberta's COVID-19 testing strategy. It was suggested that the high acceptability of saliva collection and its ability to be self-collected could be leveraged as part of the testing strategy for low-risk populations in the community. In acute care, saliva has less utility than swabs as saliva is not validated for diagnosing other respiratory infections if the test for SARS-CoV-2 is negative. There was also some discussion of the practical barriers of using saliva as a test sample in communities – anecdotal evidence suggests that sample labelling errors occur even when collected by healthcare workers, and that collection and labelling errors could be amplified by unsupervised self-collection of saliva.

Recommendations

1. NP or OP swabs are still the preferred methods of sample collection for SARS-CoV-2 diagnosis in Alberta.
Rationale: Internal data from Alberta Precision Laboratories suggests that OP and NP swabs have comparable performance in the lab-developed test used in Alberta, despite external evidence suggesting that NP swabs offer superior sensitivity to OP swabs. In addition, there are practical barriers that limit the utility of NP swabs that do not apply to OP swabs in certain settings, such as provider scope of practice, poor tolerance for NP swabbing in some populations, and availability of consumables (to a lesser degree).
2. Saliva appears to have comparable sensitivity to conventional swab techniques. Following internal validation, saliva may be used in Alberta with an appropriate collection method and protocol in an appropriate population.
Rationale: Rigorous clinical validation of the protocol for specimen collection and handling will be necessary (and is already underway), due to the ease with which the saliva specimens are affected by individual disease intensity, activities, environmental conditions, and physical characteristics of the sample itself.

Practical Considerations

- The ongoing discussion of asymptomatic COVID-19 has been [previously reviewed by the Scientific Advisory Group](#). The findings of this review suggest that in populations with low disease prevalence, asymptomatic testing is effective for identifying 90% of SARS-CoV-2-positive individuals and may be useful for close-contact testing in outbreak control situations.
- Practical implementation of saliva testing would require development of a program for saliva specimen collection and processing in addition to technical validation. Program development and implementation should include consideration of the feasibility of the sample handling and processing pathway, the most appropriate populations for saliva testing, and the most appropriate sample collection method, while balancing the human and other resources needed to implement such a program with other COVID-19

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testing initiatives. Table 1 below presents some of the factors to consider when comparing saliva specimens to NP/OP swab specimens.

Table 1. Comparability of the practical application of using saliva or swabbing techniques to test for SARS-CoV-2 infection.

	Saliva	Nasopharyngeal Swab	Throat Swab
Validated for use in Alberta (Oct 2020)	Yes	Yes	Yes
Sample reliability	Unclear	High	High
Appropriate for testing high-risk populations (eg. healthcare settings)	No	Yes	Yes
Appropriate for testing low-risk populations (eg. community monitoring)	Potentially	No	Yes
Acceptability for patients as a collection method	High	Low	Moderate
Sample can be used to diagnose other respiratory infections	Not demonstrated	Yes	Suboptimal for most
Healthcare provider PPE requirement	Low	High	High
Availability of collection devices (e.g. swabs)	High	Moderate	Moderate
Collection limited by provider scope of practice	No	Yes	No
Appropriate for unsupervised self-collection by laypeople	Moderate	No	No
Ease of handling by lab personnel	Low	High	High

Strength of Evidence

Overall, the body of evidence is of low-moderate quality. As with much of the evidence related to COVID-19, published work on test system validation appears to be opportunistic rather than carefully planned and the biases reflect this. The primary literature is at high risk of bias, however, the number of systematic reviews and meta-analyses on the topics at hand help to mitigate the biases of the individual studies but may quickly be outdated due to the rapidly evolving nature of the evidence.

The results of the included studies are partially relevant to Alberta due to the differences in outbreak dynamics, sample collection logistics, and the in-house testing protocols. Studies comparing specimens or assays often published the concordance of the comparator to the reference standard, rather than the actual sensitivity and specificity of the method under scrutiny. This method offers some evidence as to the quality of the comparator test or specimen but is only independently useful if the standard used in Alberta is equivalent to that of the research group. The laboratories in Alberta use either a laboratory-developed real-time RT-PCR or any one of several Health Canada-authorized commercial tests. All tests have been evaluated for adequacy and, if implemented, have been found to be suitable for the diagnosis of COVID-19. Any novel specimens or test assays will need to be validated against one of the currently-used methods in Alberta (or equivalent commercial assay) to ensure appropriateness.

Limitations of this review

This review is subject to substantial limitations. There is a high risk of selection bias – samples for the validation studies were often obtained from populations with a high likelihood of COVID-19 (such as emergency departments or COVID-19 units), thus over-representing positive specimens. These strategies also systematically

exclude asymptomatic individuals or those with mild symptoms that may not present to hospital or get tested for COVID-19. The artificial high prevalence of COVID-19 in these sample sets may have skewed the sensitivity and specificity results, as the false negative rate increases as the likelihood of test positivity increases.

Specifically related to research questions 1, there were several studies that were poorly controlled and did not process their samples in equivalent ways. For example, in many studies comparing saliva with nasopharyngeal swabs, the swab would be processed immediately for diagnostic purposes while the saliva sample would be refrigerated or frozen for several hours prior to processing. Viral ribonucleic acid (RNA) is highly susceptible to degradation and the differences in specimen handling could influence the quality of RNA available for the tests to detect.

Summary of Evidence

Twenty-one articles (16 peer-reviewed) from the database search are included in the narrative summary below. Of these, 5 systematic reviews were included (1 was pre-review), 0 RCTs were included, 15 observational (prospective or cross-sectional) studies were included (4 were pre-review), 0 clinical validation studies were included, 0 commentaries were included, 0 guidelines from reputable sources were included, and 1 piece of reputable grey literature were included. Two articles were included *ad hoc* (Mohammadi et al., 2020; Berenger et al., 2020). Evidence extraction tables for each research question are included in the appendix of this report (Tables 5 & 6).

Do the testing characteristics vary between nasal, nasopharyngeal, and/or throat swabs vs. saliva samples?

Evidence from secondary and grey literature

Six reviews were identified in the literature that report on the testing characteristics of saliva compared to conventional swabbing techniques; of these, four include a meta-analysis that pools the results.

It appears saliva is a challenging specimen to use due to its variability and physical characteristics (Fakheran, Dehghannejad & Khademi, 2020; Fernandes et al., 2020; Health Information and Quality Authority (HIQA), 2020; Riccò et al., 2020). Saliva can be derived from multiple places in the oral cavity and can be affected by the intensity of disease, time of day, activities (such as smoking, eating, or drinking), collection method (eg. drooling vs. spitting vs. swabbing), and storage environment (Fernandes et al., 2020; Peeters et al., in preprint). No studies were identified that used saline gargle to collect saliva. Of note, Peeters (preprint) does not observe a significant difference in sensitivity between spitting and oral swabbing methods of saliva collection.

Accordingly, the reviews included here report a high degree of variability in saliva samples. The sensitivity of saliva samples ranged from 31% to 100% (Riccò et al., 2020; HIQA, 2020; Fernandes et al., 2020). Specificity was somewhat less variable, ranging from 71% to 100% (Riccò et al., 2020; Fernandes et al., 2020), suggesting the use of imperfect reference methods in some of the primary literature. Much of this variability is lost when the sensitivity and specificity are pooled, despite methods that included all reported sensitivities. A meta-analysis by Czumbel et al. (2020) reported a pooled sensitivity of 91% for saliva compared to 98% from NP swab; meta-analysis by Peeters (preprint) reported a pooled relative sensitivity of 0.97 for saliva, which was not significantly different from NP swabs; Riccò (2020) reported a pooled specificity estimate of 97.7% (95%CI 93.8–99.2) and a pooled sensitivity estimate of 83.4% (95% CI 73.1–90.4). In their meta-analysis, Riccò (2020) found moderate agreement between NP and salivary specimens by Cohen's kappa (0.750, 95%CI 0.62-0.88).

Evidence from the primary literature

The primary literature included here consists of studies that were not included in the meta-analyses presented above. Sensitivity, specificity, and diagnostic agreement values are presented in table 2 below. Further details of each study are included in the evidence extraction (Table 5) in the appendix.

Sensitivity values for saliva specimens are extremely variable and range from 22.4% (Mestdagh et al, preprint) to 97% (Mestdagh et al., preprint) and were reported in all included studies. Specificity values for saliva specimens are generally high and range from 89.2% (Güçlü et al., 2020) to 99.9% (Yokota et al., 2020). In studies that

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reported it, the agreement between saliva specimens and conventional swab specimens ranged from 91.3% (Altawalah et al., 2020) to 96.1% (Vaz et al., 2020). A local study comparing saliva to NP swabs collected in parallel from 75 patients found that saliva had a sensitivity of 84.1% (95%CI 73.7-90.9%) compared to 91.3% (95%CI 82.3-95.9%) when using either being positive as the reference standard (Berenger et al., 2020). This protocol directed patients to allow saliva to accumulate for 1-2 minutes, spit into an empty sterile urine container (no specific volume of saliva was mandated), and then add 3 mL of Universal Transport Medium (UTM); samples were tested immediately upon receipt in the laboratory (Berenger et al., 2020).

There was little consistency in the study methods for sample collection and processing, thus it is difficult to determine which methods (and studies) ought to be considered as “high quality”. Study participants were generally asked to refrain from smoking, eating, chewing gum, and drinking for at least 30 minutes prior to sample collection. Saliva was collected into a sterile collection vessel (Griesemer et al., preprint; Lai et al., 2020; Landry et al., 2020; Mestdagh et al., 2020; Nacher et al., preprint; Senok et al., 2020; Vaz et al., 2020; Yokota et al., 2020) or into saline or transport media (Akgun Dogan et al., preprint; Altawalah et al., 2020; Güçlü et al., 2020; Kim et al., 2020; Sujipto et al., 2020). Only Mestdagh (2020) also collected saliva by oral swabbing. As described above, sample processing also plays a role in the utility of saliva specimens. Many studies did not describe if samples were processed on demand or if they were held. Senok (2020) processed samples on demand; refrigeration (4°C) was used by five studies (Akgun Dogan et al., preprint; Griesemer et al., preprint; Nacher et al., preprint; Sutjipto et al., 2020; Yokota et al., 2020); and -80°C was used by two studies (Landry et al., 2020; Vaz et al., 2020).

Table 2. Clinical testing characteristics of saliva. Unless otherwise noted, the saliva specimens were paired with conventional nasal, nasopharyngeal, or throat swabs. The reference standard was the conventional specimen, tested using the standard diagnostic RT-PCR assay for SARS-CoV-2 in use in that jurisdiction. Full details on each study are available in table 5 in the appendix.

Reference	Study size (% positive for SARS-CoV-2)	Sensitivity	Specificity	Overall agreement
Akgun Dogan et al., preprint	200 (49% positive)	63% (Day 0) 55% (Day 5)	-	-
Altawalah et al., 2020	891 (39% positive)	83.43% (95% CI: 79.07–87.20)	96.71% (95% CI: 94.85–98.04%),	91.25% ($\kappa = 0.814$, 95% CI 0.775–0.854)
Berenger et al., 2020	75 (unclear)	84.1% (95%CI 73.7-90.9%) (NP reference) 91.3% (95%CI 82.3-95.9%) (Any positive reference)	-	-
Chong et al., 2020	20 (unclear)	46.7% (Day 1-3) 52.9% (Day 4-7) 25% (Day 8-10) 33.3% (Day 11-15)	-	-
Griesemer et al., preprint	227 (41% positive)	87.1% (95% CI: 79.57-93.55)	-	-
Güçlü et al., 2020	64 (unclear)	85.2%	89.2%	$\kappa = 0.774$ ($p < 0.001$)
Kim et al., 2020	15 (100% positive)	64%	-	-
Lai et al., 2020	50 (79% positive)	84.2%	98.9%	-
Landry et al., 2020	124 (26.6% positive)	85.7% (95% CI: 70.6%–93.7%)	-	94.4% ($\kappa = 0.851$ (95 % CI 0.745 to 0.958)

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Reference	Study size (% positive for SARS-CoV-2)	Sensitivity	Specificity	Overall agreement
Mestdagh et al., preprint	2500 (4% positive)	Spitting (low viral load): 30.8%; (CI= 22.5%-40.6%)	-	-
		Swabbing (low viral load): 22.4%; (CI=15.2%-31.7%)		
		Spitting (high viral load): 97.0%; (CI=82.4%-99.8%)		
		Swabbing (high viral load): 76.7%; (CI=57.3%-89.4%)		
Nacher et al., preprint	776 (21% positive)	<10 days after symptom onset: 77%	-	-
		Ct <30: 83%; 88.9% for two genes		
Senok et al., 2020	401 (8.7% positive)	73.1% (95% CI 52.2–88.4%)	97.6% (95% CI 95.5–98.9%),	96.0% (95% CI 93.6–97.7%) κ = 0.68 (95% CI 0.53–0.82)
Sutjipto et al., 2020	105 (70% positive)	38-52%	-	-
Vaz et al., 2020	155 (43% positive)	94.4% (95% CI 86.4–97.8)	97.6% (95% CI 91.7 – 99.3),	96.1% κ = 0.922, (95% CI 0.765–1.00)
Yokota et al., 2020	1924 (2.9%)	92% (90% CI: 83-97%)	99.9% (90%CI: 99.9-100.0%)	Probability of concordance: 0.934 to 0.999; prevalence 0-30%

A recent systematic review and meta-analysis by Mohammadi et al. (2020) of 3442 respiratory samples found that in hospitalized patients, sputum had the highest percentage of positive results (71%), compared to 54% for NP swabs and 43% for OP swabs. These values were highest for the first seven days following symptom onset (Mohammadi et al., 2020). A study performed at APL using parallel NP and OP swab collections in 77 patients found that, using either result being positive as the reference standard, the positive agreement was 88.4% (95%CI 73.3-94.4%) for NP swabs and 89.9% (95%CI 77.5-93.6%) for OP swabs; in the same study, nasal swabs had a positive agreement of 80% (95%CI 62.7-90.5%) when compared to NP and OP swabs (n=36) (Byron Berenger, personal communication). Studies included here that compared saliva to nasopharyngeal and oropharyngeal swabs (Table 3) often treated them interchangeably, with the overall sensitivity of the two swabbing methods ranging from 60% on day 5 post symptom onset (Akgun Dogan et al., in preprint) to 97.8% (Greisemer et al., in preprint). Greisemer et al. (preprint) was the only study to include nasal swabs as a comparator – they found identical sensitivity as saliva (87%).

Table 3 below shows the comparative sensitivities of saliva compared to swab specimens in studies that reported the characteristics of both samples. On balance, it appears that conventional swabbing methods are superior to saliva for test sensitivity; however, the sensitivity of saliva is not so inferior that a validated collection and testing protocol would not have a place in the Alberta COVID-19 testing model. There is no recommended level of

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accuracy for COVID-19 testing – it varies by jurisdiction, risk tolerance, and logistical challenges of the testing method.

Table 3. Sensitivity of saliva specimens compared to conventional sample collection methods for SARS-Cov-2 testing, where comparisons were made. Reference method varied for each study and was not necessarily the NP swab. Full details on each study are available in table 5 in the appendix.

Reference	Study size (% positive for SARS-CoV-2)	Sensitivity of saliva	Sensitivity of swab (method)	Sensitivity of other specimen, if available
Akgun Dogan et al., preprint	200 (49% positive)	63% (Day 0) 55% (Day 5)	83% (OP/NP) Day 0 60% (OP/NP) Day 5	-
Griesemer et al., preprint	227 (41% positive)	87.1% (79.6-93.6%)	97.8% (NP)	87.1% (79.6-93.6%) (Nasal swab) 94.6 (Nasal swab + Saliva)
Kim et al., 2020	15 (100% positive)	64%	74% (NP/OP)	68% (Sputum)
Landry et al., 2020	124 (26.6% positive)	85.7% (95% CI: 70.6%–93.7%)	94.3 % (95 % CI 81.4%–99.0%) (NP)	-
Nacher et al., preprint	776 (21% positive)	<10 days after symptom onset: 90% Ct <30: 83%; 88.9% for two genes	“Saliva was less sensitive than NP swabs”	-
Sutjipto et al., 2020	105 (70% positive)	38-52%	85% (NP) 80% (Throat) 62% (Mid-turbinate)	-
Yokota et al., 2020	1924 (2.9% positive)	92% (90% CI: 83-97%)	86% (90% CI: 77-93%) (NP)	-

Does the sensitivity and specificity of SARS-CoV-2 non-serologic tests (ie. molecular or antigen tests) vary between asymptomatic or symptomatic patients?

Evidence from the primary literature

The difference in testing characteristics between symptomatic and asymptomatic COVID-19 cases is poorly reported in the literature. In general, it appears that test specimens collected from asymptomatic individuals are less sensitive than those collected from symptomatic individuals. The sensitivity of tests for asymptomatic and symptomatic individuals are presented in Table 4 below. With the exception of Yokota et al. (2020), the sensitivity of specimens obtained from asymptomatic individuals is substantially lower than that of specimens collected from symptomatic individuals. Yokota (2020) found that in a population with low disease prevalence (such as contacts of a case or travel screening populations), test sensitivity in asymptomatic individuals high (90%) in both saliva and nasopharyngeal swab samples. They tested this assumption by calculating concordance probability for prevalence from 0-30%; concordance only varied between from 0.93-0.99 (Yokota et al., 2020). More information about each study is available in Table 5 in the appendix.

Table 4. Sensitivity of tested specimens collected from symptomatic and asymptomatic individuals. Full details on each study are available in table 6 in the appendix.

Reference	Study size (number asympt)	Symptomatic sensitivity	Asymptomatic sensitivity
Holborow et al., 2020	172 (85)	86% (throat swab)	67% (throat swab)

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Mestdagh et al., preprint	2500 (705)	34.6%; CI=22.3%-49.2% (saliva, spitting device)	13.3%; CI=4.4%-31.6% (saliva, both devices)
		26.9%; CI=16.0%-41.3% (saliva, swabbing device)	
Nacher et al., preprint	776 (302)	90% (NP)	24% (NP swab)
Yokota et al., 2020	1924 (1924)	n/a	90% (both NP and saliva)

Evolving Evidence

The evidence on this topic is rapidly evolving – research groups publish opportunistically as novel testing methods are adopted or considered. The evidence presented here is a useful starting point for the discussions regarding implementing novel specimen collection. The evidence should be revisited in the future (3-6 months) to address advances in the field.

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Authorship and Committee Members

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Appendix

Evidence Extraction Tables

Table 5. Evidence extraction table for research question 1 (Difference in testing characteristics between saliva and conventional swabbing methods)

Reference	Study type	Study description	Findings	Notes
Akgun Dogan et al., in preprint	Cross-sectional study (n=200); Turkey	<ul style="list-style-type: none"> - Saliva (1 ml), nasopharyngeal and oro-nasopharyngeal samples collected from 200 consecutive patients presenting to hospital on day 0 (within 24 hours of presentation) and day 5 - Before the saliva collection, participants were given brief explanations about the difference between saliva and sputum, then they were asked to give saliva samples prior to other samples by a drooling technique. They spit approximately 1 mL into the falcon tubes containing the viral transport medium (VTM). - All samples were transferred to our laboratory within 1 hour of sampling, stored in the refrigerator, and RT-PCR was performed on the day they were collected for <i>ORF1ab</i> and <i>N</i> genes 	<ul style="list-style-type: none"> - 98 of 200 (49%) patients in the study group were positive in RT-PCR analysis performed on the samples taken as a standard diagnostic procedure before the hospitalization - Day 0: The sensitivity rate was observed as 55/66 (83%) for both oro-nasopharyngeal and nasopharyngeal samples, while it was 35/66 (63%) for saliva samples ($p < 0.001$) (of 66 samples with any positive sample) - Day 0: of 102 patients with a previous negative result, 7 tested positive in at least one sample; ONP positive in all 7 samples, NP positive in 3 samples, Saliva positive in 1 sample. - Day 5: The sensitivity rate was determined as 11/20 (55%) for both saliva and nasopharyngeal samples, while it was 12/20 (60%) for oro-nasopharyngeal samples (n.s.) (of 20 samples with any positive sample) 	<ul style="list-style-type: none"> - Unclear when “day 0” occurs relative to symptom onset - Saliva is significantly less sensitive in earlier samples than NP or OP; this difference does not persist to day 5
Altawalah et al., 2020	Cross-sectional study (n=891); Kuwait	<ul style="list-style-type: none"> - Paired saliva (~1.5 ml) and NP samples obtained from 891 consecutively admitted patients suspected of COVID-19 - Whole saliva (≈1.5 mL) was collected after deep cough from the suspected patients into a sterile container. Viscous saliva was added to 300 μL of viral transport media (VTM), mixed vigorously, and then 200 μL of sample was used for RNA isolation - Samples tested by RT-PCR for <i>Orf1ab</i>, <i>N</i>, and <i>S</i> genes - The result was considered positive if cycle threshold (Ct) values were <37 for three SARS-CoV-2 targets (the <i>ORF1ab</i>, the <i>N</i>, and the <i>S</i> genes). Samples positive for one or two targets were considered equivocal 	<ul style="list-style-type: none"> - Of the 891 suspected subjects, 38.61 % (344/891) were positive for SARS-CoV-2, 4.83 % (43/891) were equivocal, and 56.56 % (504/891) were negative with NPS by RT-PCR - For saliva, 34.23 % (305/891) were positive for SARS-CoV-2, 3.14 (28/891) were equivocal, and 62.63 % (558/891) were negative - No significant difference between NP and saliva for negative or equivocal specimens - Saliva had significantly higher detection of negative samples ($p < 0.01$) - Using NPS RT-PCR as the reference standard, the sensitivity and specificity of RT-PCR for the diagnosis of COVID-19 in saliva were 83.43 % (95 % CI: 79.07–87.20) and 96.71 % (95 % CI: 94.85–98.04 %), respectively. - 91.25 % observed agreement (κ coefficient = 0.814, 95 % CI, 0.775–0.854). 	<ul style="list-style-type: none"> - Unclear when testing occurs relative to symptom onset - Saliva is not significantly different from NP sample collection for positive samples - Unclear sample handling procedure

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Reference	Study type	Study description	Findings	Notes
Chong et al., 2020	Letter (cross-sectional study) n= 18; China	<ul style="list-style-type: none"> - Paired NP and saliva (0.5 ml) specimens from children admitted to hospital - 18 included children; 66.7% asymptomatic, 33.3% symptomatic - tested for <i>E</i> gene by RT-PCR 	<ul style="list-style-type: none"> - In 5 (27.8%) patients, saliva PCR was persistently negative, including 1 asymptomatic child who only had samples tested on day 6 of admission (NP Ct 37.9, saliva negative). In another 5 (27.8%) patients, saliva that was initially negative on day 1-3 turned positive on day 4-7 - Saliva PCR had higher Ct compared to NP swabs. The Ct differences were statistically significant for all time periods except day 11-15 - Peak saliva sensitivity was 52.9% compared to NP swabs. 46.7% (Day 1-3) 52.9% (Day 4-7) 25% (Day 8-10) 33.3% (Day 11-15) 	<ul style="list-style-type: none"> - 7 out of 55 (12.7%) paired samples had a delayed (3-28 hours) first saliva collection while awaiting NP confirmation of COVID-19. - Unclear when testing occurs relative to symptom onset - Unclear collection method and sample handling
Czumbel et al., 2020	Systematic review (n=26) and meta-analysis (n=5)	<ul style="list-style-type: none"> - Search includes records from 1 Jan 2020 – 25 April 2020 - We included records if they met the following eligibility criteria: (1) records published in scientific journals or clinical trial registries; (2) patients diagnosed with COVID-19; (3) index test: saliva specimens with PCR diagnostics for detecting SARS-CoV-2; (4) reference standard (comparator test): NPS specimens with PCR diagnostics for detecting SARS-CoV-2; (5) records written in English or available in English translation. - 26 articles included in qualitative synthesis; 5 articles included in meta-analysis 	<ul style="list-style-type: none"> - No significant difference in SARS-CoV-2 detection sensitivity between saliva and NPS specimens (moderate heterogeneity) - the test sensitivities for SARS-CoV-2 were 91% (CI 80–99%) and 98% (CI 89–100%) for saliva and for NPS samples, respectively, based the pooled event rates among COVID-19 patients - Moderate risk of bias in both individual studies and overall 	Only two studies describing specificity of saliva tests; these are not pooled in this analysis
Fakheran, Dehghannejad & Khademi, 2020	Scoping review (n= 9 studies)	<ul style="list-style-type: none"> - Systematic literature search - Studies selected for review included original, full-text articles published in English, evaluating saliva as diagnostic specimen for detecting COVID-19 patients. All letters, narrative reviews, animal studies, and duplicate articles were excluded - 9 studies included in qualitative synthesis 	<ul style="list-style-type: none"> - Most of the studies included in this review reported that there is no statistically significant difference between nasopharyngeal or sputum specimens and saliva samples regarding viral load - Method of saliva collection and device is critical for using saliva as an effective clinical specimen - Saliva collection has advantages in clinical practice – it is non-invasive 	- No meta-analysis; studies used in this review are included here
Fernandes et al., 2020	Systematic review (n= 28 studies)	<ul style="list-style-type: none"> - Systematic literature search - Studies were eligible for inclusion if they assessed the potential diagnostic value or other discriminatory properties of biological markers in the saliva of patients with COVID-19. Studies were excluded if they were 1) 	<ul style="list-style-type: none"> - Saliva samples were collected in different ways, such as cough saliva, posterior oropharyngeal saliva, saliva swab, and unstimulated saliva. The most commonly used term was saliva, without detailing the sample collection technique. No study directly compared those types of sampling 	- It appears that while the testing characteristics of saliva are comparable to conventional collection methods, saliva appears

Reference	Study type	Study description	Findings	Notes
		not original research (reviews), 2) conference abstracts, 3) written in non-Latin alphabet, or 4) not peer-reviewed. - 28 articles included in qualitative synthesis	- Nine studies reported the sensitivity and/or specificity of RT-qPCR-analyzed saliva specimens as compared with the gold standard diagnosis of throat and nasopharyngeal swabs, which varied considerably from 66% to 91.7% and from 97% to 100%, respectively. - Studies retrieved from this review reported that the sensitivity of RT-qPCR-analyzed saliva specimens was 66% to 92% for COVID-19 as compared with the standard diagnosis with throat and nasopharyngeal swabs	to be less consistent, as it appears to be affected by the intensity of disease, collection time and method, PCR protocol, and storage environment
Griesemer et al., in preprint	Comparative study (n= 227); New York	- Matched NPS, nasal swab (NS), and saliva specimens collected from community testing sites (41% positive) - Nasal swab and NPS were placed in separate tubes containing 1 ml Molecular Transport Media - Saliva samples were collected in sterile 50mL conical tubes, and patients were instructed to refrain from eating, drinking, chewing gum or tobacco, or smoking, 30 minutes prior to collection - Specimens were held at 4°C from the time of collection to the time of processing into lysis buffer for molecular testing. All testing was performed within 24-72 hours of the time of specimen collection - Validation samples tested on CDC 2019 nCoV Real-Time RT-PCR Diagnostic Panel	- NPS and the combination of NS and saliva provided the highest sensitivities (97.8% and 94.6%, respectively) with overlapping 95% confidence intervals - Both NS and saliva had lower, and identical, sensitivities and 95% confidence intervals (87.1% and 79.57-93.55 for both - By combining the NS and saliva specimens in the laboratory, we were able to increase sensitivity to 95%, an additional 8% above NS or saliva alone; we believe this to be a substantial improvement and a beneficial option for sensitive diagnosis of SARS-CoV-2	- Unclear symptom status of participants
Güçlü et al., 2020	Cross-sectional study (n=64); Turkey	- Paired Oro-nasopharyngeal (ONS) and saliva samples. The swab was placed into a 5 ml tube containing 2 ml viral transport medium (VTM). The patients were asked to collect the saliva sample themselves. They were given a sterile dry container and told to close the lid of the container after placing the saliva in it. - Samples tested with Genesis RT-PCR SARS-CoV-2 kit Patients divided into three groups: - Group 1 (30 patients): Hospitalized patients with a finding consistent with COVID-19 in the CT scan of the lung and detected SARS-CoV-2 by PCR in at least one ONS sample. Samples collected on day 3 of hospitalization	- In 23 (35.9%) of the patients, both saliva and ONS samples were positive at the same time, in 4 (6.25%) patients, only the saliva, and in another 4 (6.25%) patients, only the ONS was positive. SARS-CoV-2 was detected in the saliva samples of 27 (42.2%) patients - Saliva testing characteristics: Sensitivity: 85.19% Specificity: 89.19% PPV: 85.19% NPV: 89.19% - SARS-CoV-2 was detected in the saliva samples of 27 (42.2%) patients. The value of kappa was substantial in agreement as 0.744 and it was found to be statistically significant (<0.001)	- Unclear when samples collected relative to symptom onset - Poor sample processing detail

Reference	Study type	Study description	Findings	Notes
		<ul style="list-style-type: none"> - Group 2 (15 patients): Hospitalized patients with a finding compatible with COVID-19 in lung CT examination, but in whom SARS-CoV-2 were not detected in at least two ONS samples by PCR. Samples collected on day 3 of hospitalization - Group 3 (19 patients): Patients who present to the emergency department with complaints compatible with COVID-19 (fever, cough, shortness of breath) but have normal CT. Samples collected at admission. 		
Health Information and Quality Authority, 2020	Evidence summary (Grey literature)	- Rapid evidence review comparing nasopharyngeal, oropharyngeal and lower respiratory tract samples with salivary samples	<ul style="list-style-type: none"> - For suspected SARS-CoV-2 cases, positive detection by the comparators of interest to this review ranged from 79.3% to 100%; detection by saliva ranged from 64.7% to 100%. Positive agreement between samples for overall detection ranged from 57.4% to 100%. Negative agreement between samples ranged from 72.7% to 100%. - For known SARS-CoV-2 infected cases, positive detection by the comparators of interest to this review ranged from 41.9% to 100%; detection by saliva ranged from 30.8% to 100%. Positive agreement of detection between samples ranged from 30.8% to 100%. 	
Kim et al., 2020	Prospective study (n= 15); South Korea	<ul style="list-style-type: none"> - Matched NP/OP swab, saliva (1-2 ml), and sputum samples collected at day 1 of admission and every day until two sequentially negative tests - In patient's room, 1–2 mL of saliva or sputum was collected in sterile containers, and then diluted using same volume of sterile saline or universal transport medium in the Laboratory. - 15 patients with SARS-Cov-2 infection - RNA amplified using PowerChek™ 2019-nCoV Real-time PCR Kit for <i>RdRP</i> and <i>E</i> genes 	<ul style="list-style-type: none"> - Overall sensitivity of rRT-PCR using saliva was 64% (34/53), which is lower than the 77% (41/53) using NP/OP swabs - The sensitivities of rRT-PCR using NP/OP swabs, sputum, and saliva were 74% (23/31), 68% (21/23) and 71% (22/31) in patients with sputum - The sensitivity of rRT-PCR using saliva (8/15, 53%) was especially significantly lower than that using the NP/OP swab specimen (14/15, 93%) in early stage (1–5 days after symptom onset; P = 0.013) 	<ul style="list-style-type: none"> - Authors suggest that saliva is not appropriate for initial diagnosis of COVID-19 and shouldn't replace NP/OP swabs - Unclear if samples were processed on demand or held for a period of time.
Lai et al., 2020	Prospective study (n=50); Hong Kong	<ul style="list-style-type: none"> - Matched serial conventional respiratory tract specimens including sputum and pooled NP and throat swabs. - Patients first cleared their throat by gargling with their own saliva, and then they spit out the DTS into a sterile bottle. Sputum samples were self-collected. Patients were asked to cough out sputum and spit into a sterile plastic bottle 	<ul style="list-style-type: none"> - The overall RT-PCR-positive rate for all specimen types combined was 79.2% (446 of 563) - DTS showed the lowest RT-PCR-positive rate per individual patient compared with those of sputum and pooled NP and throat swabs (mean positive rate of DTS = 72.3%, 95% confidence interval [CI] = 62.6%–81.8%; mean positive rate of sputum = 91.7%, 95% CI = 83.8%–99.6%; 	<ul style="list-style-type: none"> - Unclear where in the care pathway participants were recruited from (eg. inpatient, emergency, etc.)

Reference	Study type	Study description	Findings	Notes
		<ul style="list-style-type: none"> - Early morning samples before tooth brushing and breakfast were preferred - RT-PCR for <i>N</i> gene using in-house test 	<ul style="list-style-type: none"> mean positive rate of pooled NP and throat swabs = 82.6%, 95% CI = 76.7%–88.6%) - Pearson correlation index showed that DTS had higher correlation to sputum (0.76; 95% CI, 0.62–0.86) than to pooled NP and throat swabs (0.67; 95% CI, 0.48–0.79) - The sensitivity and specificity of RT-PCR using saliva specimens were 84.2% and 98.9%, respectively, when using NP and throat swabs as the reference standard 	<ul style="list-style-type: none"> - Unclear if samples were processed on demand or held
Landry et al., 2020	Prospective study (n=124); Connecticut	<ul style="list-style-type: none"> - Paired NPS and saliva samples collected from 124 symptomatic participants at drive-through testing sites - Patients were asked to not eat or drink for 30 min, let saliva pool in their mouths and then spit into sterile containers. Samples were kept in a cooler and delivered within 2 h to the laboratory. - NPS samples tested immediately, residual NPS and saliva samples frozen at -70C and tested within 2 weeks - RT-PCR using CDC protocol for <i>N1</i>, <i>N2</i>, and <i>RnaseP</i> (human) 	<ul style="list-style-type: none"> - 33/124 NPS (26.6 %) were PCR positive, and saliva was also positive for 28 of these 33 (84.8 %) NPS-saliva pairs - 35 samples were RT-PCR positive, with 33/35 positive by NPS (sensitivity = 94.3 % (95 % CI 81.4%–99.0%)) and 30/35 by pure saliva (sensitivity = 85.7 % (95 % CI 70.6%–93.7%)), for an overall agreement of 117/124 (94.4 %) between the two sample types - Cohen's kappa of 0.851 (95 % CI 0.745 to 0.958) 	<ul style="list-style-type: none"> - Unclear when samples collected relative to symptom onset - Authors note difficulty working with at least a third of saliva samples
Mestdagh et al., in preprint	Prospective study (n= 2500); Belgium	<ul style="list-style-type: none"> - 2500 paired NP and saliva specimens - Saliva specimens collected by saliva swabbing device, spitting device, or both - In parallel, saliva samples were collected using Norgen Biotek's Saliva RNA Collection and Preservation Device Dx #53800 (for collection of 2 ml of saliva through spitting), DNA Genotek's ORAcollect RNA device # ORE-100 (for saliva collection through swabbing), or both. Participants in the study were asked not to eat, drink, smoke or use chewing gum in the last 30 minutes preceding saliva sampling. - - RNA extraction was performed using the Total RNA Purification Kit (Norgen Biotek #24300) according to the manufacturer's instructions using 200 µl viral transport medium (for the NP swab) or 200 µl saliva, 200 µl lysis buffer and 200 µl ethanol, with processing using a centrifuge 	<ul style="list-style-type: none"> - Out of 2884 nasopharyngeal swab samples analyzed by test lab 1, 117 (4.0%) were SARS-CoV-2 positive. There were 107/117 nasopharyngeal positive samples for which a matching saliva spitting sample was available, and 107/117 nasopharyngeal positive samples for which a matching saliva swabbing sample was available - 33/107 (sensitivity = 30.8%; CI= 22.5%-40.6%) saliva spitting samples and 24/107 (sensitivity = 22.4%; CI=15.2%-31.7%) saliva swabbing samples that were SARS-CoV-2 positive - for individuals with a high viral load (E-gene Cq < 24.5 in the nasopharyngeal sample), concordance between the nasopharyngeal and matching saliva sample improved dramatically, especially for the saliva spitting device resulting in high sensitivity in this subgroup (sensitivity = 97.0%; CI=82.4%- 99.8% and sensitivity = 76.7%; CI=57.3%-89.4% for the saliva obtained by spitting and swabbing, respectively) 	<ul style="list-style-type: none"> - Higher NP load in true-positive saliva samples than in those with false-positive saliva samples - Saliva sampling issues tested and found to be no different between collection devices - NP sampling issues may contribute to some but not all of the differences between saliva and NP specimens - Sensitivity of saliva samples was high when there was a high viral load in the NP sample, regardless of symptom status

Reference	Study type	Study description	Findings	Notes
		-RT-PCR using Charité E gene assay and in-house controls (lab 1) or TaqPath COVID-19 Combo Kit (comprising <i>ORF1ab</i> , <i>N</i> gene, and <i>S</i> gene (lab 2)	- sensitivity in saliva for SARS-CoV-2 detection was higher among symptomatic cases (sensitivity = 34.6%; CI=22.3%-49.2% and sensitivity = 26.9%; CI=16.0%-41.3% for spitting and swabbing saliva device respectively) compared to asymptomatic cases (sensitivity = 13.3%; CI=4.4%-31.6% for both the spitting and swabbing saliva device)	- Unclear if samples processed on demand or held
Nacher et al., in preprint	Prospective study (n= 776); French Guiana	- Paired NP and saliva specimens collected from 776 participants at testing tents and mobile testing brigades (162 positive by either NP or saliva) - The trained nurse present during the testing mission performed the nasopharyngeal swab and collected the salivary sputum sample in a urine container. - Samples stored at 4°C until analysis (unclear time frame) - RT-PCR using GeneFinder COVID-19 test for <i>RdRp</i> , <i>E</i> and <i>N</i> genes	- 84% had a symptoms onset <10 days, and 4% were hospitalized within 2 weeks after inclusion - For symptomatic patients for whom the interval between symptoms onset and sampling was <10 days sensitivity was 77%, but when excluding persons with isolated N gene positivity (54/162), sensitivity was 90% - In asymptomatic patients, sensitivity was 24% - For patients with Ct values <30, sensitivity was 83% or 88.9% when considering 2 genes - The positive predictive value of saliva samples was 88.4% and the negative predictive value was 88.9% - saliva testing was less sensitive than nasopharyngeal swabs	- NP is used as reference for saliva - Saliva collection could have influenced sensitivity - High number of uninfected individuals in the denominator
Peeters et al., in preprint	Systematic review and meta-analysis (n=12 studies; 1070 patients)	- 8 studies included in meta-analysis - Studies were included if subjects were tested using a RT-qPCR method detecting RNA of the SARS-CoV-2 virus on saliva samples and NP samples. Covariates of interest were: study design, severity of symptoms, hospitalization status, and timing of testing (days after onset of symptoms). We only selected paired studies, where the two types of specimen were collected from the same patients	- The sensitivity of SARS-CoV-2 testing was not significantly lower on saliva compared to testing on NP swabs (pooled relative sensitivity was 0.97, 95% CI=0.92-1.02, I ² =24%) - The relative sensitivity did not differ significantly (p=0.242) by method of saliva collection: 0.98 [95% CI=0.91-1.06] for spitting in vials and 0.94 [95% CI=0.84-1.04] for oral swabbing - Influence of disease severity on saliva sensitivity could not be assessed	- Possible influencing covariates are severity of symptoms, hospitalization status, confirmation status of COVID-19 disease at enrollment and method to collect saliva (swabbing or spitting).
Riccò et al., 2020	Systematic review and meta-analysis (n= 14 studies)	- 14 studies included in meta-analysis; 1118 samples - Only articles (a) dealing with COVID-19 cases diagnosed by means of conventional RT-qPCR tests on rhinopharyngeal swabs (5); (b) analyzing saliva by means of RT-qCPR; (c) reporting the raw number of true positive/true negative, and false positive/false negative results were eligible for the full review.	- Specificity ranged from 71.4% to 100% - The pooled estimate was 97.7% (95%CI 93.8–99.2), without significant differences between synchronous (98.0%, 95%CI 95.5 – 99.1) and diachronous studies (97.7%, 95%CI 72.6-99.0) - Sensitivity ranged from 31.3% to 100% - Pooled sensitivity was 83.4% (95%CI 73.1–90.4), resulting from 85.7% (95%CI 72.6-93.2) for diachronous studies, and 80.3% (95%CI 61.8-91.1) for synchronous studies	- High heterogeneity for both sensitivity and specificity of saliva - Forest plot inspection suggests reporting bias

Testing characteristic for novel specimens, populations, and platforms • 19

Reference	Study type	Study description	Findings	Notes
			<ul style="list-style-type: none"> - moderate diagnostic agreement between conventional NP and salivary based RT-qPCR tests (i.e. Cohen's kappa = 0.750, 95%CI 0.62-0.88) - A positive test was associated with a relatively strong evidence of disease, a negative one was associated with a reduced chance of being actually affected by SARS-CoV-2 infection. 	
Senok et al., 2020	Cross-sectional study (n= 401); United Arab Emirates	<ul style="list-style-type: none"> - Paired NP and saliva (2-4 ml) samples from 401 attendees (82% male) of a designated COVID-19 screening facility (35 positive in at least one specimen) - Saliva was collected using sterile containers without transport medium and samples were obtained at least one hour after the patient last consumed food, fluid, or smoked tobacco. Patients were asked to pool saliva in their mouth for 1–2 minutes and then gently spit 2–4 mL of saliva into the provided sterile container. - Samples processed immediately upon arrival at diagnostic lab - NP Swab used as reference standard - RT-PCR using NeoPlex COVID-19 kit for <i>RdRp</i> and <i>N</i> gene targets 	<ul style="list-style-type: none"> - Sensitivity and specificity of saliva was 73.1 % (95% CI 52.2–88.4%) and 97.6% (95% CI 95.5–98.9%), respectively - The PPV and NPV were 67.9% (95% CI 51.5–80.8%) and 98.1% (95% CI 96.5–99.0%), respectively. - The accuracy was 96.0% (95% CI 93.6–97.7%) and Kappa coefficient was 0.68 (95% CI 0.53–0.82) 	
Sutjipto et al., 2020	Cross-sectional study (clinical audit) (n=105); Singapore	<ul style="list-style-type: none"> - Matched samples (NP, midturbinate, throat, saliva) collected from a convenience sample of suspected or confirmed cases of COVID-19 - To collect saliva samples, patients were asked to rinse their mouth with plain water at least 30 minutes postmeal and 10 minutes precollection to remove residual food debris. Two milliliters of fresh salivary sample was then spit out (drooling method) by the patient into a sterile container containing an equal amount of nucleic acid stabilization formula, and this was mixed after capping by gently inverting the container 5 times. All specimens were obtained by trained nurses and processed within 24 hours. - Reference standard true positives: patients with at least 1 positive SARS-CoV-2 result detected from any site on the day of the audit or at any time point thereafter 	<ul style="list-style-type: none"> - We included 105 patients in this evaluation, 32 of whom tested negative for SARS-CoV-2 (11 patients recovered from COVID-19 and 21 had alternate diagnoses) and 73 of whom had active SARS-CoV-2 infection - NP specimens were found to have the highest clinical sensitivity, at 85%, followed by throat, 80%, mid-turbinate, 62%, and saliva, 38%–52% - nasopharyngeal site was found to be more sensitive compared with mid-turbinate or saliva (either assay) ($P < .01$) - Combination testing from patients in their first week of COVID-19 showed the best performance, with a clinical sensitivity of 98% for either midturbinate or nasopharyngeal swabs combined with throat swabs 	- Unclear when testing occurs relative to symptom onset

Reference	Study type	Study description	Findings	Notes
		<ul style="list-style-type: none"> - RT-PCR using A*STAR Fortitude Kit; saliva samples also tested with EZ1 virus mini kit, version 2.0 for <i>N</i> and <i>Orf1ab</i> genes 		
Vaz et al., 2020	Prospective study (n= 155); Brazil	<ul style="list-style-type: none"> - 155 Healthcare workers and patients presenting with signs/symptoms of COVID-19 provided a paired NPS/OPS and saliva (2 ml) - Participants were instructed to repeatedly spit until approximately 2 ml of sample was obtained, thus avoiding mucous secretions from oropharynx or lower respiratory tract (i.e., sputum) - Samples stored at -80C until extraction (within 6 hours wherever possible) - NPS/OPS samples tested with BIOMOL OneStep/ COVID-19 Kit RT-PCR - Saliva subjected to Charité-Berlin protocol: first (screening) to amplify the <i>E</i> gene, a confirmatory and eliminatory step. The last two stages, targeted <i>RdRp</i> gene, were run in case nucleic acid was detected on screening - NPS/OPS samples used as reference standard - Absence of SARS-CoV-2 infection confirmed by serum ELISA in cases of discordant results 	<ul style="list-style-type: none"> - 149 (96.1%) had concordant results on the detection of SARS-CoV-2 RT-PCR in both specimens - All 67 participants diagnosed with COVID-19 had mild to moderate symptoms - the sensitivity and specificity of RT-PCR using saliva samples were 94.4% (95% CI 86.4–97.8) and 97.62% (95% CI 91.7 – 99.3), respectively - overall high agreement (96.1%) between the two tests (kappa coefficient 0.922, 95% CI 0.765–1.00, $p < 0.001$) - testing saliva as an alternative to NP swabs is sensitive and specific enough to be used in a routine practice 	
Yokota et al., 2020	Prospective study (n= 1924); Japan	<ul style="list-style-type: none"> - Two cohorts: “Contact Tracing (CT)” – asymptomatic persons in close contact with clinically confirmed COVID-19 patients (n=250); “Airport Quarantine (AQ)” – asymptomatic travelers arriving at Tokyo and Kansai airports (n=1818) - Paired NP and saliva specimens obtained and processed with 48 hours, stored at 4C - Saliva samples were self-collected in a sterilized 15mL polystyrene sputum collection tube at partitioned booth. - RT-PCR using Loopamp 2019-SARS-CoV-2 detection kit. All saliva samples in both cohorts were analyzed by both qRT-PCR and RT-LAMP 	<ul style="list-style-type: none"> - In the CT cohort, SARS-CoV-2 was detected in 41 NPS samples and in 44 saliva samples, of which 38 individuals had both samples test positive. 114 persons were negative in both tests, which resulted in 152 of 161 matches. - In the AQ cohort, viral RNA was detected in NPS and saliva in five and four samples, respectively, out of 1763 Individuals - The sensitivity of NPS and saliva were 86% (90% CI: 77-93%) and 92% (90% CI: 83-97%), respectively, and the specificity of NPS and saliva were 99.93% (90% CI: 99.77-99.99%) and 99.96% (90%CI: 99.85-100.00%), respectively - When the prevalence was varied from 0% to 30%, the point estimate for the true concordance probability ranged from 0.934 to 0.999 and the lower limit of the 90% CI was never below 0.9 	- Saliva is highly sensitive and specific

Table 6. Evidence extraction for research question 2 (Testing characteristics for symptomatic and asymptomatic individuals)

Reference	Study type	Study description	Findings	Notes
Holborow et al., 2020	Cross-sectional study (n= 127); Wales	<ul style="list-style-type: none"> - Results from two clusters of infection among healthcare workers in well-defined settings were analyzed using RT-PCR and convalescent antibody testing. 42 were symptomatic, of whom 25 were positive following a single swab. 85 individuals were asymptomatic; 73 were swabbed, 10 were positive and 63 were negative. Of the remaining 62 asymptomatic negative individuals, five were positive for SARS-CoV-2 IgG antibodies, 41 were negative and 17 were not tested - Specimens collected from a single throat swab - No details on RT-PCR test for SARS-CoV-2 genes 	<ul style="list-style-type: none"> - Sensitivity of SARS-CoV-2 RT-PCR test in asymptomatic individuals was 67% (60% in cluster 1, 80% in cluster 2) - In symptomatic individuals, sensitivity of SARS-CoV-2 RT-PCR test was 86% (87.5% in cluster 1, 85% in cluster 2) 	
Mestdagh et al., in preprint	Prospective study (n= 2500); Belgium	<ul style="list-style-type: none"> - 2500 paired NP and saliva specimens - Saliva specimens collected by saliva swabbing device, spitting device, or both - RT-PCR using Charité E gene assay and in-house controls (lab 1) or TaqPath COVID-19 Combo Kit (comprising <i>ORF1ab</i>, <i>N</i> gene, and <i>S</i> gene (lab 2) 	<ul style="list-style-type: none"> - Out of 2884 nasopharyngeal swab samples analyzed by test lab 1, 117 (4.0%) were SARS-CoV-2 positive. There were 107/117 nasopharyngeal positive samples for which a matching saliva spitting sample was available, and 107/117 nasopharyngeal positive samples for which a matching saliva swabbing sample was available. - The fraction of subjects that were SARS-CoV-2 positive in the nasopharyngeal sample was similar in the symptomatic and asymptomatic group, 4.67% and 4.96% respectively - For 2172 study participants, we were able to register presence or absence of symptoms for COVID-19. From these, 1412 (65.0%) were symptomatic, 705 (32.5%) were asymptomatic and 55 (2.5%) indicated they experienced symptoms in the 2 weeks preceding the test - Sensitivity in saliva for SARS-CoV-2 detection was higher among symptomatic cases (sensitivity = 34.6%; CI=22.3%-49.2% and sensitivity = 26.9%; CI=16.0%-41.3% for spitting and swabbing saliva device respectively) compared to asymptomatic cases (sensitivity = 13.3%; CI=4.4%-31.6% for both the spitting and swabbing saliva device) 	<ul style="list-style-type: none"> - Higher NP load in true-positive saliva samples than in those with false-positive saliva samples - Saliva sampling issues tested and found to be no different between collection devices - NP sampling issues may contribute to some but not all of the differences between saliva and NP specimens - Sensitivity of saliva samples was high when there was a high viral load in the NP sample, regardless of symptom status

Reference	Study type	Study description	Findings	Notes
Nacher et al., in preprint	Prospective study (n= 776); French Guiana	<ul style="list-style-type: none"> - Paired NP and saliva specimens collected from 776 participants at testing tents and mobile testing brigades (162 positive by either NP or saliva) - The trained nurse present during the testing mission performed the nasopharyngeal swab and collected the salivary sputum sample in a urine container. - Samples stored at 4°C until analysis (unclear time frame) - RT-PCR using GeneFinder COVID-19 test for <i>RdRp</i>, <i>E</i> and <i>N</i> genes 	<ul style="list-style-type: none"> - 84% had a symptoms onset <10 days, and 4% were hospitalized within 2 weeks after inclusion - 39% asymptomatic - For symptomatic patients for whom the interval between symptoms onset and sampling was <10 days sensitivity was 77%, but when excluding persons with isolated N gene positivity (54/162), sensitivity was 90% - In asymptomatic patients, sensitivity was 24% - For patients with Ct values <30, sensitivity was 83% or 88.9% when considering 2 genes 	<ul style="list-style-type: none"> - NP is used as reference for saliva - Saliva collection could have influenced sensitivity - High number of uninfected individuals in the denominator
Yokota et al., 2020	Prospective study (n= 1924); Japan	<ul style="list-style-type: none"> - Two cohorts: “Contact Tracing (CT)” – asymptomatic persons in close contact with clinically confirmed COVID-19 patients (n=250); “Airport Quarantine (AQ)” – asymptomatic travelers arriving at Tokyo and Kansai airports (n=1818) - Paired NP and saliva specimens obtained and processed with 48 hours, stored at 4C - Saliva samples were self-collected in a sterilized 15mL polystyrene sputum collection tube at partitioned booth. - RT-PCR using Loopamp 2019-SARS-CoV-2 detection kit. All saliva samples in both cohorts were analyzed by both qRT-PCR and RT-LAMP 	<ul style="list-style-type: none"> - In the CT cohort, SARS-CoV-2 was detected in 41 NPS samples and in 44 saliva samples, of which 38 individuals had both samples test positive. 114 persons were negative in both tests, which resulted in 152 of 161 matches. - In the AQ cohort, viral RNA was detected in NPS and saliva in five and four samples, respectively, out of 1763 Individuals - qRT-PCR in both NP and saliva had specificity greater than 99.9% and sensitivity approximately 90% in asymptomatic patients, validating the current practice of detecting infection by nucleic acid amplification 	<ul style="list-style-type: none"> - Saliva is highly sensitive and specific

COVID-19 Scientific Advisory Group

Rapid Evidence Report

List of Abbreviations

AHS: Alberta Health Services

AQ: Airport Quarantine

CI: Confidence interval

COVID-19: Coronavirus Disease-2019

CRS: Composite Reference Standard

CT: Contact Tracing

Ct: Cycle threshold

DSO: Days from Symptom Onset

DTS: Deep-throat secretions

ED: Emergency department

EUA: Emergency Use Authorization

FDA: Food and Drug Administration

HIQA: Health Information and Quality Authority

KRS: Knowledge Resource Services

LDT: Lab-developed test

LTC: Long-term Care

NAAT: Nucleic Acid Amplification Technology

NP: Nasopharyngeal

NPA: Negative Percent Agreement

NPS: Nasopharyngeal Swab

NPV: Negative Predictive Value

NS: nasal swab

OA: Overall Agreement

OP: Oropharyngeal

OPA: Overall Percent Agreement

POC: Point of Care

POCT: Point of Care Test

PPA: Positive Percent Agreement

PPV: Positive Predictive Value

RADT: Rapid Antigen Diagnostic Test

RDT: Rapid Diagnostic Test

RNA: ribonucleic acid

RT-PCR: Reverse transcriptase polymerase chain reaction

SAG: Scientific Advisory Group

UTM: Universal Transport Media

VTM: Viral Transport Media

WHO: World Health Organization

Methods

Literature Search

A literature search was conducted by Lauren Seal and Rachel Zhao from Knowledge Resources Services (KRS) within the Knowledge Management Department of Alberta Health Services. KRS searched databases for articles published in 2020 and included: Medline, CINAHL, PubMed/LitCOVID, Trip Pro, Google Scholar, medRxiv/BioRxiv, and grey literature from CEBM, CADTH, CDC, and WHO. A separate search was conducted for each research question; the full search strategy is included in this appendix.

Articles identified by KRS in their search were initially screened by the librarian for obvious irrelevance. Articles were then screened by title and abstract against the inclusion/exclusion criteria listed in Table 7 below. In total, 271 articles were identified by KRS with references and abstracts provided for further review. Duplicates that were retrieved from different search strategies were not removed. 147 articles were excluded following title and abstract review, and an additional 104 articles were excluded following full-text screening and evidence extraction in accordance with the inclusion/exclusion criteria stated below. 50 articles were included in the final narrative synthesis.

Table 7. Inclusion and exclusion criteria for results of the literature search

Inclusion Criteria	Exclusion Criteria
<ul style="list-style-type: none"> - Any test population - COVID-19 - Includes sensitivity, specificity and/or predictive values - Q1 only: Compares saliva samples with conventional sampling (NP, throat, nasal) - Q2 only: compares tests from symptomatic and asymptomatic cases - BKit Virus Finder COVID-19 (Hyris Ltd) - BD Veritor System for Rapid Detection of SARS-CoV-2 (BD & Company) - Abbott ID NOW COVID-19 (Abbott Diagnostics Scarborough) - Panbio COVID-19 Ag Rapid Test Device (Abbott Rapid Diagnostics) - Xpert Xpress SARS-CoV-2 (Cepheid) - Human study - English language - Any jurisdiction 	<ul style="list-style-type: none"> - Article is not from a credible source - Article does not have a clear research question or issue - Presented data/evidence is not sufficient to address the research questions - Viruses other than COVID-19 - Blood sample testing - Test characteristic of samples alone (no comparison) - Serological study - Infectivity study - Analytical sensitivity - Sample pooling advice - Q1 only: Compares aspects of testing protocol OTHER than sample site or symptoms (eg. collection method, PCR type, extraction methods, etc.) - Q2 only: does not compare tests from symptomatic and asymptomatic cases

<ul style="list-style-type: none"> - Meta-analysis, Systematic review, guidelines, controlled trial, observational study, grey literature, preprint - Q4 only: commentary, narrative review - Published in 2020 	<ul style="list-style-type: none"> - Spartan Cube COVID-19 System (Spartan Bioscience Inc.) - Commentary, opinion, editorial, narrative review; modelling study
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Critical Evaluation of the Evidence

Exclusion criteria for study quality were adapted from the Mixed Methods Appraisal Tool (MMAT) (Hong et al., 2018). Potential articles were evaluated on three criteria: 1) Peer reviewed or from a reputable source; 2) Clear research question or issue; 3) Whether the presented data/evidence is appropriate to address the research question. Preprints and non peer-reviewed literature (such as commentaries and letters from credible journals) are not excluded out of hand due to the novelty of COVID-19 and the speed with which new evidence is available.

Table 8 below is a narrative summary of the body of evidence included in this review. The categories, format, and suggested information for inclusion were adapted from the Oxford Centre for Evidence-Based Medicine, the Cochrane Library, and the AGREE Trust (Urwin, Gavinder & Graziadio, 2020; Viswanathan et al, 2012; Wynants et al., 2020; Brouwers et al., 2010).

Table 8. Narrative overview of the literature included in this review.

	Description
Volume	5 systematic reviews were included (1 was pre-review), 0 RCTs were included, 15 observational (prospective or cross-sectional) studies were included (4 were pre-review), 0 clinical validation studies were included, 0 commentaries were included, 0 guidelines from reputable sources were included, and 1 piece of reputable grey literature were included.
Quality	<p>The body of evidence included in this review is of moderate quality overall. As with much of the evidence related to COVID-19, published work on test validation appears to be opportunistic rather than carefully planned and the risk of bias reflects this.</p> <p>There is a high risk of selection bias – in the observational studies, specimens were often obtained from emergency departments or COVID-19 units rather than from community testing sites. Specimens were taken from those with clinical suspicion of or confirmed COVID-19, thus ensuring an over-representation of positive specimens in the sample set. This sample collection strategy used in many studies heavily biases towards symptomatic patients, as it systematically excludes asymptomatic or paucisymptomatic individuals that may not present to hospital or get tested for COVID-19. The artificial high prevalence of COVID-19 in these sample sets may have skewed the sensitivity and specificity results, as the false negative rate increases as the likelihood of positivity increases.</p> <p>In many cases, those interpreting the test results for the sample or assay being validated were not blinded to the results obtained by the reference standard method for the correlated paired sample. In studies where concordance or agreement was measured, this has the potential to skew the results towards higher concordance than the true value.</p> <p>Specifically related to research questions 1 (saliva), there were several studies that were poorly controlled and did not process their samples in equivalent ways. For example, in many studies comparing saliva with nasopharyngeal swabs, the swab</p>

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	would be processed immediately for diagnostic purposes while the saliva sample would be refrigerated or frozen for several hours prior to processing. In several studies comparing rapid testing methods, the sample collection and storage methods were different or the samples for the comparator methods were processed differently from the reference method (eg. on demand for the reference vs. frozen residual sample for the comparator). Viral RNA is highly susceptible to degradation and the differences in specimen handling could influence the quality of RNA available for the tests to detect.
Applicability	The results of the included studies are somewhat applicable to Alberta. Studies comparing specimens or assays often published the concordance of the comparator to the reference standard, rather than the actual sensitivity and specificity of the method under scrutiny. This method offers some evidence as to the quality of the comparator test or specimen, but is only independently useful if the standard used in Alberta is the same as that of the research group. Since Alberta uses a lab-developed RT-PCR assay to test for COVID-19, any novel specimens or test assays will need to be validated against the in-house method to ensure appropriateness.
Consistency	The evidence appears to be consistent across studies.

Search Strategy

Strategy for Research Question 1

Medline

- 1 exp Coronavirus/ or Coronavirus Infections/ or coronaviru*.mp. or corona viru*.mp. or ncov*.mp. or n-cov*.mp. or novel cov*.mp. or COVID-19.mp. or COVID19.mp. or COVID-2019.mp. or COVID2019.mp. or SARS-CoV-2.mp. or SARSCoV-2.mp. or SARSCoV2.mp. or SARSCoV19.mp. or SARS-Cov-19.mp. or SARSCov-19.mp. or SARSCoV2019.mp. or SARS-Cov-2019.mp. or SARSCov-2019.mp. or severe acute respiratory syndrome coronaviru*.mp. or severe acute respiratory syndrome cov 2.mp. or 2019 ncov.mp. or 2019ncov.mp. (69105)
- 2 "severe acute respiratory syndrome*".mp. (35127)
- 3 "severe acute respiratory disease*".mp. (55)
- 4 Severe Acute Respiratory Syndrome/ (5153)
- 5 1 or 2 or 3 or 4 (72220)
- 6 Saliva/ (41877)
- 7 saliva.mp. (64675)
- 8 spit.mp. (564)
- 9 6 or 7 or 8 (65132)
- 10 Nasal Cavity/ (11681)
- 11 exp Nasal Mucosa/ (26073)
- 12 nasal.mp. (136374)

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- 13 Pharynx/ (19065)
- 14 throat.mp. (23477)
- 15 nasopharyngeal.mp. (36109)
- 16 10 or 11 or 12 or 13 or 14 or 15 (210730)
- 17 exp "Sensitivity and Specificity"/ (589412)
- 18 exp Diagnostic Errors/ (117085)
- 19 sensitivity.mp. (1176944)
- 20 specificity.mp. (1063439)
- 21 "false negative*".mp. (45716)
- 22 "true negative*".mp. (3600)
- 23 "true positive*".mp. (8514)
- 24 "false positive*".mp. (76453)
- 25 "positive predictive value*".mp. (49297)
- 26 "negative predictive value*".mp. (49116)
- 27 "test valid*".mp. (1056)
- 28 validity.mp. (182693)
- 29 validation.mp. (288024)
- 30 17 or 18 or 19 or 20 or 21 or 22 or 23 or 24 or 25 or 26 or 27 or 28 or 29 (2383654)
- 31 5 and 9 and 16 and 30 (27)

CINAHL

- S1 ((MH "Coronavirus+") OR (MH "Coronavirus Infections+") OR coronaviru* OR "corona virus" OR ncov* OR n-cov* OR ("2019 ncov" OR 2019ncov OR Hcov*)) OR (COVID-19 OR COVID19 OR COVID-2019 OR COVID2019) OR (SARS-COV-2 OR SARSCOV-2 OR SARSCOV2 OR SARSCOV19 OR SARS-COV-19 OR SARSCOV-19 OR SARSCOV2019 OR SARS-COV-2019 OR SARSCOV-2019) OR (MH "Severe Acute Respiratory Syndrome") OR (("severe acute respiratory syndrome cov 2" OR "severe acute respiratory syndrome coronavirus*") OR "severe acute respiratory syndrome" OR "severe acute respiratory disease*")
26,293
- S2 (MH "Saliva") 8,209
- S3 saliva OR spit 11,528
- S4 S2 OR S3 11,528
- S5 (MH "Nasal Cavity") 1,499
- S6 (MH "Nasal Mucosa") 1,616
- S7 nasal OR throat OR nasopharyngeal 31,305

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S8	(MH "Pharynx+")	6,348
S9	S5 OR S6 OR S7 OR S8	34,736
S10	(MH "Sensitivity and Specificity")	84,242
S11	(MH "Diagnostic Errors+")	20,399
S12	(MH "Predictive Value of Tests")	51,354
S13	sensitivity OR specificity OR "true positive*" OR "true negative*" OR "false positive*" OR "false negative*" OR "positive predictive value*" OR "negative predictive value*" OR "test valid*" OR validation OR validity	558,557
S14	S10 OR S11 OR S12 OR S13	595,864
S15	S1 AND S4 AND S9 AND S14	5

PubMed

"coronavirus"[MeSH Terms] OR "coronavirus infections"[MeSH Terms] OR "coronaviru*"[Title/Abstract] OR "corona virus*"[Title/Abstract] OR "ncov*"[Title/Abstract] OR "n cov*"[Title/Abstract] OR "novel cov*"[Title/Abstract] OR "covid-19"[Title/Abstract] OR "Covid19"[Title/Abstract] OR "covid-2019"[Title/Abstract] OR "covid2019"[Title/Abstract] OR "sars-cov-2"[Title/Abstract] OR "sarscov-2"[Title/Abstract] OR "sarscov2"[Title/Abstract] OR "sars-cov-2019"[Title/Abstract] OR "sarscov2019"[Title/Abstract] OR "sars-cov-19"[Title/Abstract] OR "sarscov19"[Title/Abstract] OR "severe acute respiratory syndrome coronaviru*"[Title/Abstract] OR "severe acute respiratory syndrome*"[Title/Abstract] OR "severe acute respiratory disease*"[Title/Abstract] OR "2019 ncov"[Title/Abstract] OR "2019ncov"[Title/Abstract] OR "severe acute respiratory syndrome"[MeSH Terms]

"saliva"[MeSH Terms] OR "saliva*"[Title/Abstract] OR "spit"[Title/Abstract]

"nasal cavity"[MeSH Terms] OR "nasal mucosa"[MeSH Terms] OR "pharynx"[MeSH Terms] OR "nasal"[Title/Abstract] OR "throat"[Title/Abstract] OR "nasopharyngeal"[Title/Abstract]

"sensitivity and specificity"[MeSH Terms] OR "diagnostic errors"[MeSH Terms] OR "sensitivity"[Title/Abstract] OR "specificity"[Title/Abstract] OR "false negative*"[Title/Abstract] OR "false positive*"[Title/Abstract] OR "true negative*"[Title/Abstract] OR "true positive*"[Title/Abstract] OR "positive predictive value*"[Title/Abstract] OR "negative predictive value*"[Title/Abstract] OR "test valid*"[Title/Abstract] OR "validity"[Title/Abstract] OR "validation"[Title/Abstract]

Trip Pro/Google Scholar

("covid-19" OR coronavirus OR COVID19 OR "corona virus" "covid-2019" OR covid2019 OR "SARS-COV-2" OR "sarscov-2" OR sarscov2 "severe acute respiratory syndrome") AND (saliva OR spit) AND (throat OR nasopharyngeal OR nasal) AND (sensitivity OR specificity OR "positive predictive value" OR "negative predictive value" OR "false positive" OR "false negative" OR "true positive" OR "true negative" OR validity OR validation OR "predictive value of tests") from:2020

("covid-19" OR coronavirus OR COVID19 OR "corona virus" "covid-2019" OR covid2019 OR "SARS-COV-2" OR "sarscov-2" OR sarscov2 "severe acute respiratory syndrome") AND (saliva OR spit) AND (throat OR nasopharyngeal OR nasal) AND (sensitivity OR specificity OR "positive predictive value" OR "negative predictive value" OR "false positive")

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("covid-19" OR coronavirus OR COVID19 OR "corona virus" "covid-2019" OR covid2019 OR "SARS-COV-2" OR "sarscov-2" OR sarscov2 "severe acute respiratory syndrome") AND ("false negative" OR "true positive" OR "true negative" OR validity OR validation OR "predictive value of tests")

medRxiv/CEBM/CADTH/CDC/WHO

("covid-19" OR coronavirus OR "corona virus" OR SARS-COV-2) AND (saliva OR spit) AND (throat OR nasal or nasopharyngeal)"

Saliva

Strategy for Research Question 2

Medline

- 1 exp Coronavirus/ or Coronavirus Infections/ or coronaviru*.mp. or corona viru*.mp. or ncov*.mp. or n-cov*.mp. or novel cov*.mp. or COVID-19.mp. or COVID19.mp. or COVID-2019.mp. or COVID2019.mp. or SARS-CoV-2.mp. or SARSCoV-2.mp. or SARSCoV2.mp. or SARSCoV19.mp. or SARS-Cov-19.mp. or SARSCov-19.mp. or SARSCoV2019.mp. or SARS-Cov-2019.mp. or SARSCov-2019.mp. or severe acute respiratory syndrome coronaviru*.mp. or severe acute respiratory syndrome cov 2.mp. or 2019 ncov.mp. or 2019ncov.mp. (69262)
- 2 "severe acute respiratory syndrome*".mp. (35138)
- 3 "severe acute respiratory disease*".mp. (55)
- 4 Severe Acute Respiratory Syndrome/ (5153)
- 5 1 or 2 or 3 or 4 (72378)
- 6 exp "Sensitivity and Specificity"/ (589412)
- 7 exp Diagnostic Errors/ (117085)
- 8 sensitivity.mp. (1177040)
- 9 specificity.mp. (1063489)
- 10 "false negative*".mp. (45718)
- 11 "true negative*".mp. (3600)
- 12 "true positive*".mp. (8514)
- 13 "false positive*".mp. (76457)
- 14 "positive predictive value*".mp. (49304)
- 15 "negative predictive value*".mp. (49122)
- 16 "test valid*".mp. (1056)
- 17 validity.mp. (182717)
- 18 validation.mp. (288072)
- 19 6 or 7 or 8 or 9 or 10 or 11 or 12 or 13 or 14 or 15 or 16 or 17 or 18 (2383831)
- 20 exp Polymerase Chain Reaction/ (449578)

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- 21 "polymerase chain reaction*".mp. (594653)
- 22 PCR.mp. (542592)
- 23 RT-PCR.mp. (142922)
- 24 RTPCR.mp. (851)
- 25 nonserologic*.mp. (26)
- 26 non-serologic*.mp. (24)
- 27 "reverse transcriptase polymerase chain reaction*".mp. (167439)
- 28 exp Antigens, Viral/ (102610)
- 29 "antigen* test*".mp. (5403)
- 30 20 or 21 or 22 or 23 or 24 or 25 or 26 or 27 or 28 or 29 (969453)
- 31 symptomatic.mp. (191214)
- 32 "showing symptom*".mp. (600)
- 33 "displaying symptom*".mp. (141)
- 34 asymptomatic.mp. (167375)
- 35 exp Asymptomatic Diseases/ (7643)
- 36 presymptomatic.mp. (4131)
- 37 paucisymptomatic.mp. (233)
- 38 Carrier State/ (21709)
- 39 carrier.mp. (264634)
- 40 non-symptomatic.mp. (912)
- 41 31 or 32 or 33 (191807)
- 42 34 or 35 or 36 or 37 or 38 or 39 or 40 (432086)
- 43 5 and 19 and 30 and 41 and 42 (32)
- 44 31 or 32 or 33 or 34 or 35 or 36 or 37 or 38 or 39 or 40 (591319)
- 45 5 and 19 and 30 and 44 (136)
- 46 45 not 43 (104)

CINAHL

S1 ((MH "Coronavirus+") OR (MH "Coronavirus Infections+") OR coronaviru* OR "corona virus" OR ncov* OR n-cov* OR ("2019 ncov" OR 2019ncov OR Hcov*)) OR (COVID-19 OR COVID19 OR COVID-2019 OR COVID2019) OR (SARS-COV-2 OR SARSCOV-2 OR SARSCOV2 OR SARSCOV19 OR SARS-COV-19 OR SARSCOV-19 OR SARSCOV2019 OR SARS-COV-2019 OR SARSCOV-2019) OR (MH "Severe Acute Respiratory Syndrome") OR (("severe acute respiratory syndrome cov 2" OR "severe acute respiratory

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	syndrome coronavirus**) OR "severe acute respiratory syndrome" OR "severe acute respiratory disease**")	
	26,605	
S2	(MH "Sensitivity and Specificity")	84,283
S3	(MH "Diagnostic Errors+")	20,408
S4	(MH "Predictive Value of Tests")	51,407
S5	sensitivity OR specificity OR "true positive**" OR "true negative**" OR "false positive**" OR "false negative**" OR "positive predictive value**" OR "negative predictive value**" OR "test valid**" OR validation OR validity	559,692
S6	S2 OR S3 OR S4 OR S5	597,025
S7	(MH "Polymerase Chain Reaction+")	46,328
S8	(MH "Antigens, Viral+")	2,852
S9	"polyermase chain reaction" OR "revere transcriptase polymerase chain reaction" OR "reverse transcription polymerase chain reaction" OR PCR OR RTPCR OR RT-PCR OR nonserologic* OR non-serologic* OR "antigen test**"	36,612
S10	S7 OR S8 OR S9	67,724
S11	(MH "Signs and Symptoms+")	675,615
S12	(MH "Carrier State")	2,727
S13	sympomatic* OR "showing symptom*" OR "displaying symptom**" OR asymptomatic OR presymptomatic OR paucisymptomatic OR carrier OR non-symptomatic	70,093
S14	S11 OR S12 OR S13	739,038
S15	S1 AND S6 AND S10 AND S14	54
S16	S1 AND S6 AND S10 AND S14 Limiters - Published Date: 20200101-20201231	41

PubMed

("coronavirus"[MeSH Terms] OR "coronavirus infections"[MeSH Terms] OR "coronaviru**"[Title/Abstract] OR "corona virus**"[Title/Abstract] OR "ncov**"[Title/Abstract] OR "n cov**"[Title/Abstract] OR "novel cov**"[Title/Abstract] OR "covid-19"[Title/Abstract] OR "Covid19"[Title/Abstract] OR "covid-2019"[Title/Abstract] OR "covid2019"[Title/Abstract] OR "sars-cov-2"[Title/Abstract] OR "sarscov-2"[Title/Abstract] OR "sarscov2"[Title/Abstract] OR "sars-cov-2019"[Title/Abstract] OR "sarscov2019"[Title/Abstract] OR "sars-cov-19"[Title/Abstract] OR "sarscov19"[Title/Abstract] OR "severe acute respiratory syndrome coronaviru**"[Title/Abstract] OR "severe acute respiratory syndrome**"[Title/Abstract] OR "severe acute respiratory disease**"[Title/Abstract] OR "2019 ncov"[Title/Abstract] OR "2019ncov"[Title/Abstract] OR "severe acute respiratory syndrome"[MeSH Terms] AND ("sensitivity and specificity"[MeSH Terms] OR "diagnostic errors"[MeSH Terms] OR "sensitivity"[Title/Abstract] OR "specificity"[Title/Abstract] OR "false negative**"[Title/Abstract] OR "false positive**"[Title/Abstract] OR "true negative**"[Title/Abstract] OR "true positive**"[Title/Abstract] OR "positive predictive value**"[Title/Abstract] OR "negative predictive value**"[Title/Abstract] OR "test valid**"[Title/Abstract] OR "validity"[Title/Abstract] OR "validation"[Title/Abstract]) AND ("polymerase chain reaction"[MeSH Terms] OR "polymerase chain reaction**"[Title/Abstract] OR "reverse transcription polymerase chain reaction**"[Title/Abstract] OR "PCR"[Title/Abstract] OR "RT-PCR"[Title/Abstract] OR "RTPCR"[Title/Abstract] OR "antigen test**"[Title/Abstract] OR "test antigen**"[Title/Abstract] OR "antigens, viral"[MeSH Terms] OR "reverse transcriptase polymerase chain reaction**"[Title/Abstract]) AND

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("symptomatic"[Title/Abstract] OR "displaying symptom*" [Title/Abstract] OR "asymptomatic"[Title/Abstract] OR "asymptomatic diseases"[MeSH Terms] OR "signs and symptoms"[MeSH Terms] OR "carrier state"[MeSH Terms] OR "presymptomatic"[Title/Abstract] OR "paucisymptomatic"[Title/Abstract] OR "carrier"[Title/Abstract])

TRIP Pro

("covid-19" OR coronavirus OR "corona virus" OR sars-cov-2) AND (sensitivity OR specificity OR "positive predictive value" OR "negative predictive value" OR "false positive" OR "false negative" OR "true positive" OR "true negative" OR validity OR validation OR "predictive value") AND (RT-PCR OR PCR OR "antigen test" OR "reverse transcriptase polymerase chain reaction" OR "polymerase chain reaction") AND (symptomatic OR "showing symptoms" OR "displaying symptoms" OR asymptomatic OR presymptomatic OR paucisymptomatic OR "no symptoms") from:2020

Google Scholar/LitCovid/WHO

("covid-19" OR coronavirus OR "corona virus" OR sars-cov-2) AND (sensitivity OR specificity OR validity OR validation OR "predictive value") AND (RT-PCR OR PCR OR "antigen test" OR polymerase chain reaction) AND (symptomatic OR asymptomatic)

medRxiv

(COVID-19 OR sars-cov-2 OR coronavirus) AND (sensitivity OR specificity) AND (test or testing OR PCR OR RT-PCR)

CEBM/CADTH/CDC

Test

Testing

Rt-pcr

PCR

Polymerase chain reaction

Antigen test

Reference List

- Akgun Dogan, O., Kose, B., Agaoglu, N. B., Yildiz, J., Alkurt, G., Kendir Demirkol, Y., Irvem, A., Dinler-Doganay, G., & Doganay, L. (2020). Does sampling saliva increase detection of SARS-CoV-2 by RT-PCR? Comparing saliva with oro-nasopharyngeal swabs. MedRxiv, pre-print, 2020.07.26.20158618. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.07.26.20158618v1.full.pdf>
- Altawalah, H., AlHuraish, F., Alkandari, W. A., & Ezzikouri, S. (2020). Saliva specimens for detection of severe acute respiratory syndrome coronavirus 2 in Kuwait: A cross-sectional study. *Journal of Clinical Virology*, 132, 104652. Retrieved from: <https://www.sciencedirect.com/science/article/pii/S1386653220303942>
- Becker, D., Sandoval, E., Amin, A., Hoff, P. De, Diets, A., Leonetti, N., Lim, Y. W., Elliott, C., Laurent, L., Grzymalski, J., & Lu, J. T. (2020). Saliva is less sensitive than nasopharyngeal swabs for COVID-19 detection in the community setting. MedRxiv, 1–15. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.05.11.20092338v2.full.pdf>
- Brouwers, M. C., Kho, M. E., Browman, G. P., Burgers, J. S., Cluzeau, F., Feder, G., Fervers, B., Graham, I. D., Grimshaw, J., Hanna, S. E., Littlejohns, P., Makarski, J., Zitzelsberger, L., & AGREE Next Steps Consortium (2010). AGREE II: advancing guideline development, reporting and evaluation in health care. *CMAJ : Canadian Medical Association journal = journal de l'Association medicale canadienne*, 182(18), E839–E842. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3001530/>
- Chong, C. Y., Kam, K.-Q., Li, J., Maiwald, M., Loo, L. H., Nadua, K. D., Tan, N. W. H., Yung, C. F., & Thoon, K. C. (2020). Saliva is not a useful diagnostic specimen in children with Coronavirus Disease 2019. *Clinical Infectious Diseases*. Retrieved from: <https://academic.oup.com/cid/advance-article/doi/10.1093/cid/ciaa1376/5905581>
- Czumbel, L. M., Kiss, S., Farkas, N., Mandel, I., Hegyi, A., Nagy, Á., Lohinai, Z., Szakács, Z., Hegyi, P., Steward, M. C., & Varga, G. (2020). Saliva as a Candidate for COVID-19 Diagnostic Testing: A Meta-Analysis. *Frontiers in Medicine*, 7, 2020.05.26.20112565. Retrieved from: <https://www.frontiersin.org/articles/10.3389/fmed.2020.00465/full>
- Fakheran, O., Dehghannejad, M., & Khademi, A. (2020). Saliva as a diagnostic specimen for detection of SARS-CoV-2 in suspected patients: A scoping review. *Infectious Diseases of Poverty*, 9(1), 100. Retrieved from: <https://idpjournal.biomedcentral.com/articles/10.1186/s40249-020-00728-w>
- Fernandes, L. L., Pacheco, V. B., Borges, L., Athwal, H. K., de Paula Eduardo, F., Bezinelli, L., Correa, L., Jimenez, M., Dame-Teixeira, N., Lombaert, I. M. A., & Heller, D. (2020). Saliva in the Diagnosis of COVID-19: A Review and New Research Directions. *Journal of Dental Research*. Retrieved from: <https://journals.sagepub.com/doi/pdf/10.1177/0022034520960070>
- Griesemer, S. B., Slyke, G. Van, Ehrbar, D., Strle, K., Yildirim, T., Centurioni, D. A., Walsh, A. C., Chang, A. K., Waxman, M. J., & George, K. St. (2020). Evaluation of specimen types and saliva stabilization solutions for SARS-CoV-2 testing. MedRxiv, 701, 2020.06.16.20133041. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.06.16.20133041v1.full.pdf>
- Güçlü, E., Koroglu, M., Yürümez, Y., Toptan, H., Kose, E., Güneysu, F., & Karabay, O. (2020). Comparison of saliva and oro-nasopharyngeal swab sample in the molecular diagnosis of COVID-19. *Revista Da Associacao Medica Brasileira* (1992), 66(8), 1116–1121. Retrieved from: https://www.scielo.br/scielo.php?script=sci_arttext&pid=S0104-42302020000801116&tling=en
- Health Information and Quality Authority. (2020). Evidence summary for accuracy of salivary samples in SARS-CoV-2 detection compared with nasopharyngeal, oropharyngeal or lower respiratory tract samples. Retrieved from: <https://www.hiqa.ie/sites/default/files/2020-06/Evidence-summary-for-salivary-detection-of-SARS-CoV-2.pdf>
- Holborow, A., Asad, H., Porter, L., Tidswell, P., Johnston, C., Blyth, I., Bone, A., & Healy, B. (2020). The clinical sensitivity of a single SARS-CoV-2 upper respiratory tract RT-PCR test for diagnosing COVID-19 using

convalescent antibody as a comparator. *Clinical Medicine*, 20(6), clinmed.2020-0555. Retrieved from: <https://www.rcpjournals.org/content/clinmedicine/early/2020/09/10/clinmed.2020-0555.full.pdf>

Hong, Quan Nha et al. (2018). The Mixed Methods Appraisal Tool (MMAT) Version 2018 for Information Professionals and Researchers'. 1 Jan. 2018 : 285 – 291. Retrieved from: http://mixedmethodsappraisaltoolpublic.pbworks.com/w/file/attach/127916259/MMAT_2018_criteria-manual_2018-08-01_ENG.pdf

Jamal, A. J., Mozafarihashjin, M., Coomes, E., Powis, J., Li, A. X., Paterson, A., Anceva-Sami, S., Barati, S., Crowl, G., Faheem, A., Farooqi, L., Khan, S., Prost, K., Poutanen, S., Taylor, M., Yip, L., Zhong, X. Z., McGeer, A. J., Mubareka, S., & Toronto Invasive Bacterial Diseases Network COVID-19 Investigators (2020). Sensitivity of nasopharyngeal swabs and saliva for the detection of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2). *Clinical infectious diseases*. ciaa848. Advance online publication. <https://doi.org/10.1093/cid/ciaa848>

Kim, S. S. E., Lee, J. Y., Lee, A., Kim, S. S. E., Park, K. H., Jung, S. I., Kang, S. J., Oh, T. H., Kim, U. J., Lee, S. Y., Kee, S. J., & Jang, H. C. (2020). Viral Load Kinetics of SARS-CoV-2 Infection in Saliva in Korean Patients: a Prospective Multi-center Comparative Study. *Journal of Korean Medical Science*, 35(31), e287. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7415999/>

Lai, C. K. C., Chen, Z., Lui, G., Ling, L., Li, T., Wong, M. C. S., Ng, R. W. Y., Tso, E. Y. K., Ho, T., Fung, K. S. C., Ng, S. T., Wong, B. K. C., Boon, S. S., Hui, D. S. C., & Chan, P. K. S. (2020). Prospective Study Comparing Deep Throat Saliva With Other Respiratory Tract Specimens in the Diagnosis of Novel Coronavirus Disease 2019. *The Journal of Infectious Diseases*, 222(10), 1612–1619.. <https://academic.oup.com/jid/article/222/10/1612/5879767>

Landry, M. L., Criscuolo, J., & Peaper, D. R. (2020). Challenges in use of saliva for detection of SARS CoV-2 RNA in symptomatic outpatients. *Journal of Clinical Virology*, 130, 104567. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7392849/>

Mestdagh, P., Gillard, M., Arbyn, M., Pirnay, J.-P., Poels, J., Hellemsans, J., Peeters, E., Hutse, V., Vermeiren, C., Boutier, M., Wever, V. De, Soentjens, P., Djebara, S., Malonne, H., Andre, E., Smeraglia, J., & Vandesompele, J. (2020). Evaluation of saliva sampling procedures for SARS-CoV-2 diagnostics reveals differential sensitivity and association with viral load. *MedRxiv*, 2020.10.06.20207902. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.10.06.20207902v1.full.pdf>

Mohammadi, A., Esmailzadeh, E., Li, Y., Bosch, R. J., & Li, J. (2020). SARS-CoV-2 Detection in Different Respiratory Sites: A Systematic Review and Meta-Analysis. *The Lancet*. Online access. Retrieved from: [https://www.thelancet.com/journals/ebiom/article/PIIS2352-3964\(20\)30278-4/fulltext](https://www.thelancet.com/journals/ebiom/article/PIIS2352-3964(20)30278-4/fulltext)

Nacher, M., Mergeay-Fabre, M., Blanchet, D., Benois, O., Pozl, T., Sainte-Rose, V., Vialette, V., Toulet, B., Moua, A., Simon, S., Guidarelli, M., Galindo, M., Biche, B., Faurous, W., Abad, F., Fahrasmene, A., Rochemont, D., Vignier, N., & Vabret, A. (2020). Prospective comparison of saliva and nasopharyngeal swab sampling for mass screening 2 for COVID-19 3. *MedRxiv*, 2020.09.23.20150961. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.09.23.20150961v1.full.pdf>

Pasomsub, E., Watcharananan, S. P., Boonyawat, K., Janchompoo, P., Wongtabtim, G., Suksuwan, W., Sungkanuparph, S., & Phuphuakrat, A. (2020). Saliva sample as a non-invasive specimen for the diagnosis of coronavirus disease 2019: a cross-sectional study. *Clinical microbiology and infection : the official publication of the European Society of Clinical Microbiology and Infectious Diseases*, S1198-743X(20)30278-0. <https://doi.org/10.1016/j.cmi.2020.05.001>

Peeters, E., Kaur Dhillon Ajit Singh, S., Vandesompele, J., Mestdagh, P., Hutse, V., Arbyn, M., Dhillon, S. K., Singh, A., Vandesompele, J., Mestdagh, P., Hutse, V., & Arbyn, M. (2020). Rapid systematic review of the sensitivity of SARS-CoV-2 molecular testing on saliva compared to nasopharyngeal swabs. *MedRxiv*, pre-print, 2020.08.05.20168716. Retrieved from: <https://www.medrxiv.org/content/10.1101/2020.08.05.20168716v1.full.pdf>

Riccò, M., Ranzieri, S., Peruzzi, S., Valente, M., Marchesi, F., Balzarini, F., Bragazzi, N. L., & Signorelli, C. (2020). RT-qPCR assays based on saliva rather than on nasopharyngeal swabs are possible but should be interpreted with caution: Results from a systematic review and metaanalysis. *Acta Biomedica*, 91(3), 1–15. Retrieved from: <https://www.mattioli1885journals.com/index.php/actabiomedica/article/view/10020/9010>

Senok, A., Alsuwaidi, H., Atrah, Y., Ayedi, O. Al, Zahid, J. Al, Han, A., Marzooqi, A. Al, Heialy, S. Al, Altrabulsi, B., Abdelwareth, L., Idaghdour, Y., Ali, R., Loney, T., & Alsheikh-Ali, A. (2020). Saliva as an alternative specimen for molecular COVID-19 testing in community settings and population-based screening. *Infection and Drug Resistance*, 13, 3393–3399. Retrieved from: <https://www.dovepress.com/saliva-as-an-alternative-specimen-for-molecular-covid-19-testing-in-co-peer-reviewed-fulltext-article-IDR>

Sutjipto, S., Lee, P. H., Tay, J. Y., Mendis, S. M., Abdad, M. Y., Marimuthu, K., Ng, O. T., Cui, L., Chan, M., Soon, M., Lin, R. T. P., Leo, Y.-S., De, P. P., Barkham, T., Vasoo, S., Ong, S. W. X., Ang, B. S. P., Lye, D. C., Lim, P. L., ... Toh, B. K. (2020). The Effect of Sample Site, Illness Duration, and the Presence of Pneumonia on the Detection of SARS-CoV-2 by Real-time Reverse Transcription PCR. *Open Forum Infectious Diseases*, 7(9), ofaa335. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7454916/>

Urwin, S; Gavinder K, Graziadio S. (2020). What prognostic clinical risk prediction scores for COVID-19 are currently available for use in the community setting? Centre for Evidence-Based Medicine. Retrieved from: <https://www.cebm.net/covid-19/what-prognostic-clinical-risk-prediction-scores-for-covid-19-are-currently-available-for-use-in-the-community-setting/>

Vaz, S. N., Santana, D. S. de, Netto, E. M., Pedroso, C., Wang, W. K., Santos, F. D. A., & Brites, C. (2020). Saliva is a reliable, non-invasive specimen for SARS-CoV-2 detection. *Brazilian Journal of Infectious Diseases*. Retrieved from: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7458056/>

Viswanathan, M., Ansari, M. T., Berkman, N. D., Chang, S., Hartling, L., McPheeters, M., ... & Treadwell, J. R. (2012). Assessing the risk of bias of individual studies in systematic reviews of health care interventions. In *Methods guide for effectiveness and comparative effectiveness reviews [Internet]*. Agency for Healthcare Research and Quality (US). Retrieved from: <https://www.ncbi.nlm.nih.gov/books/NBK91433/>

Wynants, L., Van Calster, B., Bonten, M. M., Collins, G. S., Debray, T. P., De Vos, M., ... & Schuit, E. (2020). Prediction models for diagnosis and prognosis of covid-19 infection: systematic review and critical appraisal. *BMJ*, 369. Retrieved from <https://www.bmj.com/content/369/bmj.m1328.long>

Yokota, I., Shane, P. Y., Okada, K., Unoki, Y., Yang, Y., Inao, T., Sakamaki, K., Iwasaki, S., Hayasaka, K., Sugita, J., Nishida, M., Fujisawa, S., & Teshima, T. (2020). Mass screening of asymptomatic persons for SARS-CoV-2 using saliva. *Clinical infectious diseases: an official publication of the Infectious Diseases Society of America*, ciaa1388. Advance online publication. <https://doi.org/10.1093/cid/ciaa1388>